Journal of Clinical and Translational Hepatology **2023**
DOI: 10.14218/JCTH.2022.00103S

Original Article

Community-centered Disease Severity Assessment of Metabolic Dysfunction-associated Fatty Liver Disease

Jee-Fu Huang[1,2,3,4], Pei-Chien Tsai[1], Ming-Lun Yeh[1,2,3], Chung-Feng Huang[1,2,3], Ching-I Huang[1,2], Mei-Hsuan Lee[5], Po-Yau Hsu[1,2], Chih-Wen Wang[1,3], Yu-Ju Wei[1], Po-Cheng Liang[1], Yi-Hung Lin[1], Meng-Hsuan Hsieh[1,6], Jeng-Fu Yang[1,6], Ming-Yen Hsieh[1], Tyng-Yuan Jang[1], Ming-Jong Bair[7,8], Zu-Yau Lin[1,3], Chia-Yen Dai[1,2,3,9], Ming-Lung Yu[1,3,9,10*] and Wan-Long Chuang[1,2,3*]

[1]Hepatobiliary Division, Department of Internal Medicine, Kaohsiung Medical University Hospital, Kaohsiung Medical University, Kaohsiung; [2]Graduate Institute of Clinical Medicine, Kaohsiung Medical University, Kaohsiung; [3]Faculty of Internal Medicine, College of Medicine, Kaohsiung Medical University, Kaohsiung; [4]Center for Liquid Biopsy and Cohort Research, Kaohsiung Medical University, Kaohsiung; [5]Institute of Clinical Medicine, National Yang Ming Chiao Tung University, Taipei; [6]Department of Preventive Medicine, Kaohsiung Medical University Hospital, Kaohsiung; [7]Division of Gastroenterology, Department of Internal Medicine, Taitung Mackay Memorial Hospital, Taitung; [8]Mackay Medical College, New Taipei City; [9]School of Medicine and Doctoral Program of Clinical and Experimental Medicine, College of Medicine , National Sun Yat-sen University, Kaohsiung; [10]Center of Excellence for Metabolic Associated Fatty Liver Disease, National Sun Yat-sen University, Kaohsiung

Received: 13 November 2022 | **Revised:** 25 February 2023 | **Accepted:** 18 April 2023 | **Published online:** Month 00, 2023

Abstract

Background and Aims: Disease severity across the different diagnostic categories of metabolic dysfunction-associated fatty liver disease (MAFLD) remains elusive. This study assessed the fibrosis stages and features of MAFLD between different items. We also aimed to investigate the associations between advanced fibrosis and risk factors. ***Methods:*** This multicenter cross-sectional study enrolled adults participating in liver disease screening in the community. Patients were stratified following MAFLD diagnostic criteria, to group A (395 patients) for type 2 diabetes, group B (1,818 patients) for body mass index (BMI)>23 kg/m^2, and group C (44 patients) for BMI≤23 kg/m^2 with at least two metabolic factors. Advanced fibrosis was defined as a fibrosis-4 index>2.67. ***Results:*** Between 2009 and 2020, 1,948 MAFLD patients were recruited, including 478 with concomitant liver diseases. Advanced fibrosis was observed in 125 patients. A significantly larger proportion of patients in group C (25.0%) than in group A (7.6%) and group B (5.8%) had advanced fibrosis (p<0.01). Logistic regression analysis found that hepatitis B virus (HBV)/hepatitis C virus (HCV) coinfection (odds ratio [OR]: 12.14, 95% confidence interval [CI]: 4.04–36.52; p<0.01), HCV infection (OR: 7.87, 95% CI: 4.78–12.97; p<0.01), group C (OR: 6.00, 95% CI: 2.53–14.22; p<0.01), and TC/LDL-C (OR: 1.21, 95% CI: 1.06–1.38; p<0.01) were significant predictors of advanced fibrosis. ***Conclusions:*** A higher proportion of lean MAFLD patients with metabolic abnormalities had advanced fibrosis. HCV infection was significantly associated with advanced fibrosis.

Citation of this article: Huang JF, Tsai PC, Yeh ML, Huang CF, Huang CI, Lee MH, *et al*. Community-centered Disease Severity Assessment of Metabolic Dysfunction-associated Fatty Liver Disease. J Clin Transl Hepatol 2023. doi: 10.14218/JCTH.2022.00103S.

Keywords: Metabolic dysfunction-associated fatty liver disease; Fibrosis-4 index; Advanced fibrosis; Community screening; Viral hepatitis.

Abbreviations: ALT, alanine transaminase; AST, aspartate transaminase; BMI, body mass index; CI, confidence interval; FIB-4, fibrosis-4 index; FLI, fatty liver index; FPG, fasting plasma glucose; HBV, hepatitis B virus; HCV, hepatitis C virus; HDL-C, high-density lipoprotein cholesterol; hs-CRP, high-sensitivity C-reactive protein; LDL-C, low-density lipoprotein cholesterol; MAFLD, metabolic dysfunction-associated fatty liver disease; MetS, metabolic syndrome; NAFLD, nonalcoholic fatty liver disease; NASH, nonalcoholic steatohepatitis; OR, odds ratio; T2DM, type 2 diabetes mellitus; TC, total cholesterol; TG, triglycerides; UA, uric acid.

***Correspondence to:** Wan-Long Chuang and Ming-Lung Yu, Hepatobiliary Division, Department of Internal Medicine, Kaohsiung Medical University Hospital, 100 Tzyou Road, Kaohsiung. ORCID: https://orcid.org/0000-0002-2376-421X (WLC) and https://orcid.org/0000-0001-8145-1900 (MLY). Tel: +886-7-312-1101 ext. 7475, Fax: +886-7-312-3955, E-mail: wloch@cc.kmu.edu.tw (WLC) and fish6069@gmail.com (MLY)

Introduction

Nonalcoholic fatty liver disease (NAFLD) is the most common liver disorder globally with a prevalence of 25%[1]. The incidence has been rapidly progressing in the past several decades throughout the Asia-Pacific region in parallel with the rapid Westernization of the region[2,3]. Despite a significantly lower body mass index (BMI) and lower rates of obesity compared to other ethnic groups, Asians have a significant prevalence of NAFLD as well as other metabolic disorders such as hypertension, type 2 diabetes mellitus (T2DM), and metabolic syndrome (MetS)[4]. Extensive investigation of metabolic liver diseases with complex mechanisms is essential for diagnosis, management, and outcome prediction.

Recently, metabolic dysfunction-associated fatty liver disease (MAFLD) has been proposed as a new definition for patients with fatty liver disease[5]. A recent meta-analysis

showed that the overall prevalence of MAFLD was 38.8%. It estimated that 5.37% of lean and 29.78% of nonobese individuals had MAFLD[6]. The prevalence reached nearly half of the population in some regions[7]. The new definition was designed to avoid stigma and achieve alignment with other liver diseases, focusing on metabolic alterations of the disease and an overarching approach for disease awareness and the management of patients[8]. The major intent of the new nomenclature was to shift toward a diagnosis of inclusion based on the presence of metabolic dysfunction and hepatic steatosis. Therefore, clarification of the new definition according to disease outcome is essential and informative for early diagnosis and prevention efforts in addition to implementation of a region-based strategy. Nevertheless, disease severity in MAFLD has rarely been investigated in a community-based setting in the Asia-Pacific region.

Liver fibrosis is the major determinant and the significant predictor of long-term outcome in patients with NAFLD. There is a dose-dependent association between the risk of mortality and the stage of fibrosis, in which a higher risk of mortality is associated with a higher stage of fibrosis[9,10]. Nonalcoholic steatohepatitis patients with advanced fibrosis had a significantly higher risk of all-cause mortality and liver-related mortality compared with NASH patients without fibrosis[11]. Moreover, the risk of liver-related mortality increased on an exponential rather than linear scale with an increase in fibrosis stage[12]. Liver biopsy is an expensive invasive procedure with potential complications, a high rate of sampling error, and interobserver variability[13]. Recently, the noninvasive fibrosis-4 index (FIB-4) was validated to provide an accurate prediction of liver fibrosis and liver-related events in patients with NAFLD[11,12]. The serum-based algorithm has been adapted by major societies as a clinically useful tool for advanced fibrosis assessment[14–16]. Recently it has been also validated in patients with MAFLD with different BMIs[17–19]. Therefore, its application in a community level deserves investigation.

Taiwan is an endemic for hepatitis B virus (HBV) and hepatitis C virus (HCV) infection, and nearly half of the adults have NAFLD. This unique background provides an excellent opportunity for elucidation of the characteristics of MAFLD and the interaction between steatosis *per se* and viral infections. Consequently, we conducted a community-based study aiming to elucidate the features and characteristics of MAFLD patients. We also aimed to elucidate the disease severity between different MAFLD characteristics and the impact of the prevalent viral hepatitis on the disease severity of MAFLD.

Methods

Study population

The Ethics Committee of the Kaohsiung Medical University Hospital (Kaohsiung City, Taiwan) approved this cross-sectional study before it was initiated. The recruited subjects had participated in a multipurpose integrated health examination that was part of a nonprofit community care program at 10 primary care stations in southern Taiwan between January 2009 and December 2020. Written informed consent was obtained from patients prior to enrollment, the study interview, medical record review, anthropomorphic measurements, and blood testing. Patients who had a weekly ethanol intake of more than 140 g were excluded. Anthropometric data, which included blood pressure, waist circumference, and body weight and height, were measured by standardized techniques. The enrolled subjects fasted overnight for 12 h fast before blood tests, including high-sensitivity C-reactive protein (hs-CRP), fasting plasma glucose (FPG), insulin, total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), triglycerides (TG), uric acid (UA), and transaminase (aspartate aminotransferase [AST]/alanine aminotransferase [ALT]) levels.

Fatty liver and fibrosis assessment

Abdominal sonography was performed for each participant by well-experienced and licensed hepatologists at the same institution to ensure interobserver consistency. The precise recruitment of patients with fatty liver was further validated by the fatty liver index (FLI) on diagnosis of fatty liver by sonography.

$$\mathrm{FLI}=\frac{e^{0.953\times\log_e \text{triglycerides}+0.139\times\mathrm{BMI}+0.718\times\log_e\mathrm{GGT}+0.053\times\text{waist circumference}-15.745}}{1+e^{0.953\times\log_e\text{triglycerides}+0.139\times\mathrm{BMI}+0.718\times\log_e\mathrm{GGT}+0.053\times\text{waist circumference}-15.745}}\times 100.$$

FLI≥60 was used to rule in patients with hepatic steatosis[20]. The blood fibrosis test FIB-4 was calculated as age * AST[IU/L)]/[platelets (10^9/L) * ALT (IU/L)$^{1/2}$].[21] A cutoff of >2.67 was defined as high risk of advanced fibrosis stage.[22] The risk for advanced fibrosis was classified as low (FIB-4≤1.3) and indeterminate (1.3<FIB-4≤2.67).

MAFLD definition

We defined MAFLD as the presence of metabolic risk factors in the setting of hepatic steatosis based on the diagnostic criteria proposed by an international expert panel[23]. MAFLD was diagnosed as the presence of hepatic steatosis with ≥1 of the followings: T2DM, overweight or obese (BMI>23 kg/m^2), and the presence of at least two metabolic risk abnormalities[24]. The metabolic risk abnormalities included seven items: (1) blood pressure ≥130/85 mmHg or specific drug treatment; (2) waist circumference ≥90 cm for men and ≥80 cm for women; (3) fasting plasma TG≥150 mg/dL or specific drug treatment; (4) plasma HDL-C<40 mg/dL for men and <50 mg/dL for women or specific drug treatment; (5) prediabetes with FPG 100–125 mg/dL or hemoglobin A1c 5.7–6.4%; (6) homeostasis model assessment of insulin resistance (HOMA-IR) ≥2.5; and (7) plasma hs-CRP>2 mg/L. HOMA-IR was calculated as FPG (mg/dL) × fasting insulin level (μU/mL) / 405. We stratified the subjects to group A with T2DM, group B with BMI>23 kg/m^2, and group C with BMI≤23 kg/m^2 with at least two metabolic factors.

Statistical analysis

Pearson's chi-square test was used to compare the differences between categorical variables, and Student's *t*-test analysis/analysis of variance were performed to test differences between/among continuous variables. Logistic regression analysis was used to test the statistical significance (p<0.05) of age, sex, metabolic factors, BMI, excessive alcohol use, smoking, T2DM, and viral hepatitis markers of MAFLD by univariate or multivariate models based on clinical relevance. Two sensitivity analyses were used to validate the results by different periods and randomized assignment to testing and validation groups. The two analyses included stratification of the recruited patients by different periods and 1:1 randomization into testing and validation groups by triple bootstrap sampling. p<0.05 was considered statistically significant. Quality control procedures, database processing, and statistical analysis were performed with SAS Enterprise Guide (SAS Institute Inc., Cary, NC, USA).

Results

Patient characteristics

A total of 5,180 adults≥20 years of age participated the com-

Table 1. Baseline characteristics of the MAFLD patients

Characteristics	Total, *n*=1,948	FIB-4≤2.67, *n*=1,823	FIB-4>2.67, *n*=125	*p*-value
Age (years)	51.5±13.4	50.7±13.1	62.9±12.1	<0.01*
Females	1,015 (52.1)	949 (52.1)	66 (52.8)	0.87
Waist circumference (cm)	93.9±11.2	94.0±11.3	93.5±10.5	0.63
>90 cm for male, >80 cm for female	1,502 (77.7)	1,409 (77.8)	93 (76.2)	0.69
BMI (kg/m^2)	29.4±4.5	29.5±4.5	28.0±4.4	0.001*
Alcohol (*n*=1,572)	406 (20.8)	383 (21.0)	23 (18.4)	0.49
Smoking (*n*=1,577)	425 (21.8)	402 (22.1)	23 (18.4)	0.34
Viral infections				
HBsAg+	275 (14.1)	253 (13.9)	22 (17.6)	<0.01*
Anti-HCV+	183 (9.4)	140 (7.7)	43 (34.4)	
Both+	20 (1.0)	15 (0.8)	5 (4.0)	
Hypertension	605 (37.3)	555 (36.4)	50 (51.0)	<0.01*
Dyslipidemia	427 (27.1)	399 (27.0)	28 (28.9)	0.69
TG (mg/dL)	201.0±189.7	202.6±192.7	177.8±137.5	0.03*
TC (mg/dL)	206.2±42.5	207.6±42.4	186.4±38.4	<0.01*
HDL-C (mg/dL)	49.6±13.1	49.6±12.9	49.6±15.5	0.97
LDL-C (mg/dL)	117.9±37.3	119.3±37.2	96.7±33.2	<0.01
TC/LDL-C	1.9±1.0	1.9±0.8	2.3±2.3	0.09
T2DM	258 (13.2)	223 (15.5)	23 (23.7)	0.03*
FPG (mg/dL)	106.0±45.0	105.9±45.5	106.8±36.7	0.81
HbA1c (%)	6.3±1.4	6.3±1.4	6.5±1.6	0.42
5.7–6.4	424 (40.3)	397 (40.0)	27 (46.6)	0.56
≥6.5	279 (26.5)	264 (26.6)	15 (25.9)	
HOMA-IR	4.1±5.6	4.2±5.8	3.5±2.8	0.30
≥2.5	231 (60.6)	219 (61.5)	12 (48.0)	0.18
AST (U/L)	35.3±30.0	31.6±16.4	89.8±83.8	<0.01*
ALT (U/L)	41.4±31.9	39.4±28.4	69.8±56.8	<0.01*
Platelet (× 10^3/μL)	265.7±77.0	273.6±71.9	150.4±53.4	<0.01*

Data are means±standard deviation or *n* (%). ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; FIB-4, fibrosis-4 index; FPG, fasting plasma glucose; HbA1c, hemoglobin A1c; HBsAg+, hepatitis B surface antigen-positive; HCV, hepatitis C virus; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, homeostasis model assessment of insulin resistance; LDL-C, low-density lipoprotein cholesterol; T2DM, type 2 diabetes mellitus; TC, total cholesterol; TG, triglycerides.

munity screening. We excluded 3,232 adults who had missing laboratory data (hepatitis B surface antigen [HBsAg] in 74 subjects and anti-HCV in 78 subjects) or incomplete sonographic examination (3,080 subjects). Finally, a total of 1,948 community-based MAFLD patients (mean age of 51.5±13.4 years and 52.1% women) were enrolled. The patients had an FLI>60, and fatty liver was diagnosed by abdominal ultrasound. The mean BMI was 29.4±4.5 kg/m^2. The presence of past history for T2DM, either diagnosed previously or under antidiabetic treatment, was 258 patients. An additional 137 patients met the diagnostic criteria of T2DM during surveillance, yielding a T2DM prevalence of 20.3% (395/1,948). The study pool included 478 (24.5%) patients with concomitant liver diseases, including 275 (14.1%) HBsAg+ patients, 183 (9.4%) anti-HCV+ patients, and 20 (1%) HBsAg+ and anti-HCV+ patients. Totally there were 395 (20.3%) patients in group A, 1,818 (93.3%) in group B, and 44 (2.3%) in group C, respectively.

Disease severity among groups and the impact of viral hepatitis infections

One hundred twenty-five (6.4%) patients had advanced fibrosis confirmed by the FIB-4 value (>2.67). Those patients were older (62.9±12.1 vs. 50.7±13.1 years of age; $p<0.01$), and had lower BMIs (28.0±4.4 vs. 29.5±4.5 kg/m^2; $p=0.001$) and higher prevalence of anti-HCV+ (34.4% vs. 7.7%; $p<0.01$) than their counterparts. Their mean AST and ALT levels were also significantly higher than those without advanced fibrosis. In addition, they had lower TG, TC, LDL-C, and platelet levels compared with patients with FIB-4≤2.67 (Table 1). There was a significantly higher percentage of group C patients (25.0%, 11/44) with advanced

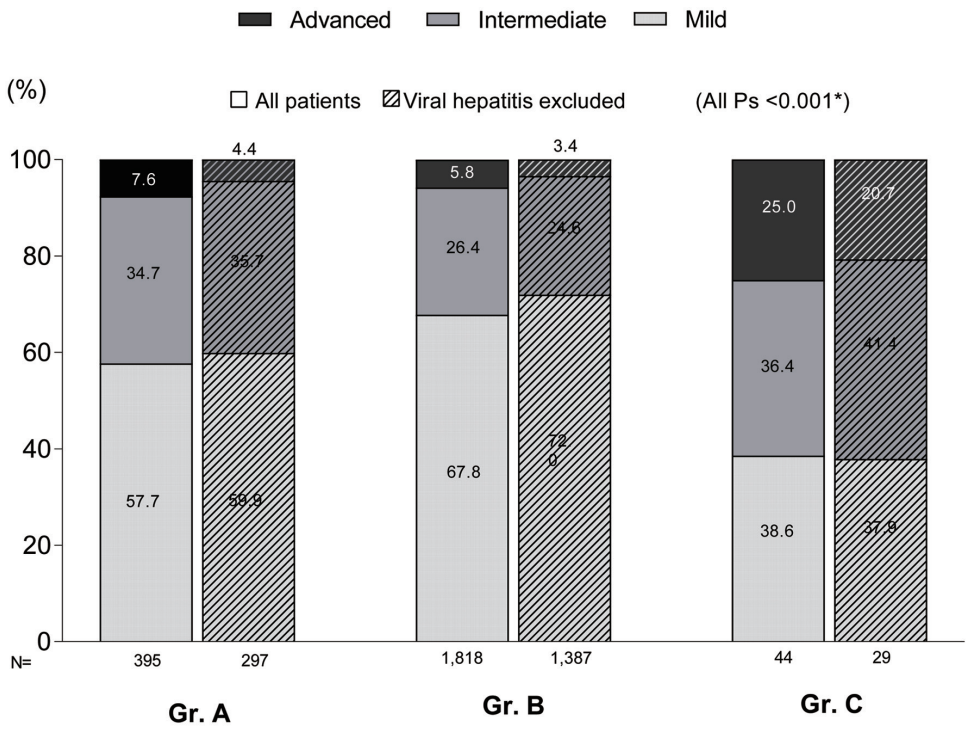

Fig. 1. Distribution of fibrosis stage. Patients with T2DM (Gr. A); patients with BMI> 23 kg/m^2 ; patients with BMI≤ 23 kg/m^2 and have at least two metabolic factors (Gr. C). BMI, body mass index; T2DM, type 2 diabetes mellitus.

fibrosis than group A (7.6%, 30/395) and group B (5.8%, 106/1,818) patients (p<0.01; Fig. 1). To determine the roles of common viral infections in MAFLD disease severity among groups, we further analyzed the results by stratifying for the presence of HBV or HCV infection. Excluding the 478 patients with concomitant viral hepatitis infections, 20.7% (6/29) of group C patients had advanced fibrosis, which was significantly higher than the proportion of group A (4.4%) and group B (3.4%) patients (p<0.01).

Associated factors for predicting advanced fibrosis

We performed multivariate logistic regression analysis to elucidate the factors associated with advanced fibrosis in MAFLD patients. The results demonstrated that HBV/HCV coinfection was the leading factor associated with advanced fibrosis (odds ratio [OR]: 12.14, 95% confidence interval [CI]: 4.04–36.52; p<0.01). The other significant factors for predicting advanced fibrosis included HCV infection (OR: 7.87, 95% CI: 4.78–12.97; p<0.01), group C (OR: 6.00, 95% CI: 2.53–14.22; p<0.01), and TC/LDL-C (OR: 1.21, 95% CI: 1.06–1.38; p<0.01; Table 2).

Validation of the associated factors for advanced fibrosis

We used two sensitivity analyses to validate the results. The first was to stratify the recruited patients by different periods. The results were consistent between the study periods of 2009–2014 and 2015–2020 (Supplementary Table 1). Group C patients, besides HCV infection and age, remained the significant factors associated with advanced fibrosis. The concordant results demonstrated that group C patients, in addition to HBV and HCV, were significant predictors of MAFLD with advanced fibrosis by triple bootstrap sampling for sensitivity analysis. (Supplementary Table 2).

Discussion

The change from NAFLD to MAFLD is more than a simple change to the nomenclature, with many clinical implications. Accordingly, the optimal approach is to elucidate the distribution and characteristics of disease severity within and between the defined components of MAFLD. Our results demonstrated that the diagnostic criteria of being overweight, namely having a BMI>23 kg/m^2, was the main cause of MAFLD in a community-based cohort. Patients with advanced fibrosis were older in age and had a lower BMI, higher TC/LDL-C, higher HCV prevalence, and a different metabolic profile than their counterparts. Of note was that there were significantly more group C patients, defined as BMI≤23 kg/m^2 with at least two metabolic factors, with advanced fibrosis (25.0%) than the other two groups. The observation remained significant after excluding the factor of viral hepatitis. Multivariate regression analysis showed that group C criteria were the major predictive factor of advanced fibrosis. Thus, our results provide evidence of a link between metabolic alterations and disease severity in MAFLD, and also shed light on the risk stratification and high-risk surveillance of metabolic liver disorders, at least at the community level.

Liver fibrosis is the process of formation and deposition of fibrous connective tissue and extracellular matrix leading to progressive structural tissue remodeling. It is a sequela of necroinflammation and/or cellular insults. A multinational, retrospective analysis of NAFLD patients demonstrated that long-term prognosis and survival after liver transplant depended less on a diagnosis of NASH or non-NASH than on the presence of fibrosis, indicating that fibrosis is the major determinant for long-term outcomes in NAFLD patients[9]. Generally, fibrosis measurement is essential for determination of the disease course and outcome of viral hepatitis infection, before and after viral eradication or sufficient suppression.

Table 2. Multivariate logistic regression analysis of risk factors predicting advanced fibrosis in MAFLD patients

Factor	FIB-4		Crude		Adjusted	
	≤2.67, *n*=1,823	>2.67, *n*=125	OR (95% CI)	*p*-value	OR (95% CI)	*p*-value
Age (years)	50.7±13.1	62.9±12.1	1.08 (1.06–1.10)	<0.01*	–	–
Female	949 (93.5)	66 (6.5)	1.03 (0.71–1.48)	0.87	1.05 (0.66–1.66)	0.83
Male	874 (93.7)	59 (6.3)	1		1	
BMI	29.5±4.5	28.0±4.4	0.92 (0.88–0.96)	<0.01*	–	–
Alcohol, Yes	383 (94.3)	23 (5.7)	0.85 (0.53–1.35)	0.49	–	–
No	1,440 (93.4)	102 (6.6)	1		–	–
Smoking, Yes	402 (94.6)	23 (5.4)	0.80 (0.50–1.27)	0.34	–	–
No	1,421 (93.3)	102 (6.7)	1		–	–
TC/LDL-C	1.9±0.8	2.3±2.3	1.21 (1.06–1.42)	<0.01*	1.21 (1.06–1.38)	<0.01*
HbA1c (%)	6.3±1.4	6.5±1.6	1.08 (0.90–1.26)	0.38	–	–
MAFLD phenotype						
BMI≤23 kg/m^2 and ≥2 metabolic items	33 (75.0)	11 (25.0)	5.38 (2.64–10.95)	<0.01*	6.00 (2.53–14.22)	<0.01*
≤23 kg/m^2 and <2 metabolic items	73 (91.3)	7 (8.7)	1.54 (0.70–4.45)	0.28	0.77 (0.22–2.68)	0.68
>23 kg/m^2	1,712 (94.2)	106 (5.8)	1		1	
T2DM						
Yes	365 (92.4)	30 (7.6)	1.26 (0.82–1.93)	0.29	0.99 (0.56–1.72)	0.96
No	1,457 (93.9)	95 (6.1)	1		1	
HBsAg+/anti-HCV+	15 (75.0)	5 (25.0)	8.58 (3.01–24.44)	<0.01*	12.14 (4.04–36.52)	<0.01*
Anti-HCV+	140 (86.5)	43 (23.5)	7.90 (5.11–12.21)	<0.01*	7.87 (4.78–12.97)	<0.01*
HBsAg+	253 (92.0)	22 (8.0)	2.24 (1.34–3.73)	<0.01*	1.02 (0.42–2.45)	0.97
HBsAg–/anti-HCV–	1,415 (96.3)	55 (3.7)	1		1	

Data are means±standard deviation or *n* (%). BMI, body mass index; FIB-4, fibrosis-4 index; HbA1c, hemoglobin A1c; HBsAg, hepatitis B surface antigen; HCV, hepatitis C virus; LDL-C, low-density lipoprotein cholesterol; MAFLD, metabolic dysfunction-associated fatty liver disease; T2DM, type 2 diabetes mellitus; TC, total cholesterol.

However, fibrosis measurement is much more vital in the context of MAFLD as there is no reliable surrogate biomarker for this complex metabolic disorder.

Several noninvasive tests to assess liver fibrosis have been developed. The FIB-4 index is a simple and easy-to-access test index with high predictive performance and reproducibility. A score of >2.67 was defined as advanced-stage fibrosis and allowed avoiding liver biopsy examination[25]. Recent practice guidelines recommend FIB-4 as the initial noninvasive test for risk stratification in NAFLD patients based on metabolic risk factors owing to its simplicity and ease of use[25,26]. However, the use of the FIB-4 index for fibrosis assessment has rarely been investigated in MAFLD in a community-based study. Our results demonstrated that a low proportion (6.4%) of MAFLD patients had advanced fibrosis as assessed by the FIB-4 index. There was a significantly higher proportion of patients with advanced fibrosis in group C than their counterpart groups. The high proportion remained significant even after excluding the factor of viral hepatitis infection. The results were validated and confirmed by sensitivity analysis using period stratification and triple bootstrap sampling. The observation was in accord with previous studies showing that metabolic abnormality is the key driver of fatty liver disease, irrespective of BMI[27,28]. Previous studies have indicated that patients with lean NAFLD had a lower prevalence of T2DM, hypertension, dyslipidemia, and MetS but higher fibrosis scores than their non-lean counterparts[29–31]. Our results suggest the benefits of high-risk surveillance of advanced fibrosis in MAFLD on group C patients, at least in a community-based approach. However, discordant results from a recent Asian study showed that the prevalence of advanced fibrosis in MAFLD was higher in group B (9.5%) than group C (3.1%) patients based on magnetic resonance elastography[32]. The discrepancy might be attributed to the differences in patient selection, the diagnostic methods for initial recruitment, fibrosis assessment tools, genetic predispositions, or racial difference[31,33,34]. Collaborative longitudinal studies across different regions and races with a uniform study design may be informative.

Recent studies consistently demonstrated that MAFLD and NAFLD do not define the same condition and should not be regarded as synonymous despite the many overlaps between the two nomenclatures[35]. Nonetheless, the new definition of MAFLD opened a wide scope for addressing the mutual impact between steatosis and viral hepatitis infection. As anticipated, age and viral infections were the major risk factors associated with advanced fibrosis. Liver steatosis is a common phenomenon in community health center patients. It is estimated that one-third of those patients have steatosis that differs in extent, possibly because of changes in host metabolism or infection with HCV genotype 3. Our results are consistent with previous studies showing a link between HCV infection and steatosis[36,37]. By contrast, the link between HBV and steatosis remains unknown. Our recent study showed that steatosis-chronic hepatitis B patients had a lower 10-year cumulative rate of cirrhosis and hepatocellular carcinoma, and a higher HBsAg seroclearance rate than their nonsteatosis counterparts[38]. HBsAg seropositivity was associated with a lower risk of developing NAFLD in a large-scale Asian study, suggesting a possible effect of HBV infection on the pathogenesis of NAFLD development[39]. A future longitudinal cohort study is needed to clarify the issue of viral-metabolic interaction from genetic to epigenetic aspects. Although the new definition is a steatosis-centered diagnosis, our study suggests the importance of surveillance for viral hepatitis in MAFLD patients.

Our study had some limitations. First, the cross-sectional design did not provide sufficient information regarding the changes in disease severity assessment in a longitudinal manner. A call-back follow-up study will be informative of differences in long-term outcome. Second, we did not use other noninvasive methods such as imaging-based modalities for measurement validation and the potential discordance between sonography and FLI. Nevertheless, the enrolled MAFLD patients were assessed by both abdominal ultrasound and FLI prior to FIB-4 evaluation for disease severity. FLI is an acceptable alternative for the diagnosis of steatosis whenever imaging tools are not available or feasible[40]. The stringent diagnosis could have decreased potential bias because MAFLD is a heterogenous and complex disorder. Third, the main clinical utility of FIB-4 in NAFLD patients lies in the ability to exclude, but not identify, advanced fibrosis[41]. Therefore, patients with indeterminate FIB-4 value might have advanced fibrosis[42]. Further validation study with histopathological approach will be helpful to improve the limitation. Lastly, we did not analyze the potential therapeutic effects of antidiabetes drugs and lifestyle modifications as this was a cross-sectional study. Currently there is no approved drug for the amelioration of fibrogenesis in MAFLD, which might greatly decrease potential bias in this aspect.

In conclusion, the study described the characteristics of disease severity in a community-based setting. Viral hepatitis infection was the major factor contributing to the occurrence of advanced fibrosis. A significantly higher proportion of lean patients with metabolic abnormalities had advanced fibrosis than their counterparts. This observation remained significant after excluding viral hepatitis infection. Further longitudinal studies are needed for risk stratification and precision prevention.

Acknowledgments

The authors are grateful for secretarial help from the Taiwan Liver Research Foundation (TLRF) and Fatty Liver Special Interest Group of Taiwan Association for the Study of the Liver (TASL). Neither the Foundation nor the Association influenced how the study was conducted or the approval of the manuscript. The study was presented in part (FRI118) at the 2022 The Internal Liver Congress of The European Association for the Study of the Liver (EASL), June 22–26, 2022.

Funding

This study was supported in part by grants from The Ministry of Science and Technology, Taiwan (MOST 110-2314-B-03-073-MY3), The Ministry of Health and Welfare, Taiwan (MOHW, 112-TDU-B-221-124007), National Yang Ming Chiao Tung University-Kaohsiung Medical University Joint Research Project (NYCU-KMU-111-I001, NYCU-KMU-112-I001), and Kaohsiung Medical University Hospital (KMUH SA10907, KMUH110-0R05).

Conflict of interest

Jee-Fu Huang: Consultant of Roche, BMS, Gilead, Merck, Sysmex, Pharmaessential, Polaris, Aligos, and Instylla. Speaker for Abbvie, BMS, Gilead, Merck, Sysmex, and Roche. Editorial board member of *Journal of Clinical and Translational Hepatology* since 2022. Chia-Yen Dai: Consultant of Abbvie and Roche; Speaker for Abbvie, Gilead, and Roche. Chung-Feng Huang: Speaker for Abbvie, BMS, Bayer, Gilead, Merck, and Roche. Ming-Lung Yu: Research grant from Abbott, BMS, Merck, and Gilead; Consultant of Abbvie, Abbott, Ascletis, BMS, Merck, Gilead, and Roche; Speaker for Abbvie, Abbott, BMS, Merck, Gilead, and IPSEN. Editorial board member of

Journal of Clinical and Translational Hepatology since 2023. Wan-Long Chuang: Consultant of Gilead, AbbVie, BMS, PharmaEssentia, and Aligos; Speaker for Gilead, AbbVie, BMS, and PharmaEssentia. Editorial board member of *Journal of Clinical and Translational Hepatology* since 2022. The other authors have no conflict of interests related to this publication.

Author contributions

Conception and design: JFH, MLY, MLY, CFH, MHL, WLC. Acquisition of data: JFH, CYD, CFH, MLY, CIH, MHH, YHL, JFY, MJB, PYH, CWW, YJW, PCL, YHL, TYJ, ZYL. Data analysis and interpretation: JFH, PCT, CIH, CFH, WLC, MLY. Manuscript drafting and critical revising: JFH, PCT, MLY, MLY, WLC. The corresponding authors attest that all listed authors meet authorship criteria. All authors provided critical review and approved the final version.

Ethical statement

The trial was conducted in compliance with the Declaration of Helsinki and the Good Clinical Practice guidelines of the International Conference on Harmonization. The study has been approved by the Institutional Review Board, Kaohsiung Medical University Hospital (KMUHIRB-E(I)-20210355).

Data sharing statement

All data relevant to the study are included in the article. Inquiries regarding the datasets used and/or analyzed during the current study can be directed to the corresponding authors.

References

[1] Younossi Z, Anstee QM, Marietti M, Hardy T, Henry L, Eslam M, *et al*. Global burden of NAFLD and NASH: trends, predictions, risk factors and prevention. Nat Rev Gastroenterol Hepatol 2018;15(1):11–20. doi:10.1038/nrgastro.2017.109, PMID:28930295.

[2] Lu SN, Wang LY, Chang WY, Chen CJ, Su WP, Chen SC, *et al*. Abdominal sonographic screening in a single community. Gaoxiong Yi Xue Ke Xue Za Zhi 1990;6(12):643–646. PMID:2266568.

[3] Hsiao PJ, Kuo KK, Shin SJ, Yang YH, Lin WY, Yang JF, *et al*. Significant correlations between severe fatty liver and risk factors for metabolic syndrome. J Gastroenterol Hepatol 2007;22(12):2118–2123. doi:10.1111/j.1440-1746.2006.04698.x, PMID:18031368.

[4] Wong RJ, Chou C, Sinha SR, Kamal A, Ahmed A. Ethnic disparities in the association of body mass index with the risk of hypertension and diabetes. J Community Health 2014;39(3):437–445. doi:10.1007/s10900-013-9792-8, PMID:24276618.

[5] Eslam M, Newsome PN, Sarin SK, Anstee QM, Targher G, Romero-Gomez M, *et al*. A new definition for metabolic dysfunction-associated fatty liver disease: An international expert consensus statement. J Hepatol 2020;73(1):202–209. doi:10.1016/j.jhep.2020.03.039, PMID:32278004.

[6] Chan KE, Koh TJL, Tang ASP, Quek J, Yong JN, Tay P, *et al*. Global Prevalence and Clinical Characteristics of Metabolic-associated Fatty Liver Disease: A Meta-Analysis and Systematic Review of 10 739 607 Individuals. J Clin Endocrinol Metab 2022;107(9):2691–2700. doi:10.1210/clinem/dgac321, PMID:35587339.

[7] Yilmaz Y, Yilmaz N, Ates F, Karakaya F, Gokcan H, Kaya E, *et al*. The prevalence of metabolic-associated fatty liver disease in the Turkish population: A multicenter study. Hepatol Forum 2021;2(2):37–42. doi:10.14744/hf.2021.2020.0033, PMID:35783905.

[8] Méndez-Sánchez N, Bugianesi E, Gish RG, Lammert F, Tilg H, Nguyen MH, *et al*. Global multi-stakeholder endorsement of the MAFLD definition. Lancet Gastroenterol Hepatol 2022;7(5):388–390. doi:10.1016/S2468-1253(22)00062-0, PMID:35248211.

[9] Angulo P, Kleiner DE, Dam-Larsen S, Adams LA, Bjornsson ES, Charatcharoenwitthaya P, *et al*. Liver Fibrosis, but No Other Histologic Features, Is Associated With Long-term Outcomes of Patients With Nonalcoholic Fatty Liver Disease. Gastroenterology 2015;149(2):389–97.e10. doi:10.1053/j.gastro.2015.04.043, PMID:25935633.

[10] Sanyal AJ, Van Natta ML, Clark J, Neuschwander-Tetri BA, Diehl A, Dasarathy S, *et al*. Prospective Study of Outcomes in Adults with Nonalcoholic Fatty Liver Disease. N Engl J Med 2021;385(17):1559–1569. doi:10.1056/NEJMoa2029349, PMID:34670043.

[11] Laryea M, Watt KD, Molinari M, Walsh MJ, McAlister VC, Marotta PJ, *et al*. Metabolic syndrome in liver transplant recipients: prevalence and association with major vascular events. Liver Transpl 2007;13(8):1109–1114. doi:10.1002/lt.21126, PMID:17663411.

[12] Dulai PS, Singh S, Patel J, Soni M, Prokop LJ, Younossi Z, *et al*. Increased risk of mortality by fibrosis stage in nonalcoholic fatty liver disease: Systematic review and meta-analysis. Hepatology 2017;65(5):1557–1565. doi:10.1002/hep.29085, PMID:28130788.

[13] Huang JF, Hsieh MY, Dai CY, Hou NJ, Lee LP, Lin ZY, *et al*. The incidence and risks of liver biopsy in non-cirrhotic patients: An evaluation of 3806 biopsies. Gut 2007;56(5):736–737. doi:10.1136/gut.2006.115410, PMID:17440193.

[14] European Association for the Study of the Liver (EASL), European Association for the Study of Diabetes (EASD), European Association for the Study of Obesity (EASO). EASL-EASD-EASO Clinical Practice Guidelines for the management of non-alcoholic fatty liver disease. Diabetologia 2016;59(6):1121–1140. doi:10.1007/s00125-016-3902-y, PMID:27053230.

[15] Chalasani N, Younossi Z, Lavine JE, Charlton M, Cusi K, Rinella M, *et al*. The diagnosis and management of nonalcoholic fatty liver disease: Practice guidance from the American Association for the Study of Liver Diseases. Hepatology 2018;67(1):328–357. doi:10.1002/hep.29367, PMID:28714183.

[16] Wong VW, Chan WK, Chitturi S, Chawla Y, Dan YY, Duseja A, *et al*. Asia-Pacific Working Party on Non-alcoholic Fatty Liver Disease guidelines 2017-Part 1: Definition, risk factors and assessment. J Gastroenterol Hepatol 2018;33(1):70–85. doi:10.1111/jgh.13857, PMID:28670712.

[17] Park H, Yoon EL, Kim M, Lee J, Kim JH, Cho S, *et al*. Comparison of diagnostic performance between FIB-4 and NFS in metabolic-associated fatty liver disease era. Hepatol Res 2022;52(3):247–254. doi:10.1111/hepr.13737, PMID:34841632.

[18] Chen X, Goh GB, Huang J, Wu Y, Wang M, Kumar R, *et al*. Validation of Non-invasive Fibrosis Scores for Predicting Advanced Fibrosis in Metabolic-associated Fatty Liver Disease. J Clin Transl Hepatol 2022;10(4):589–594. doi:10.14218/JCTH.2021.00311, PMID:36062270.

[19] Eren F, Kaya E, Yilmaz Y. Accuracy of Fibrosis-4 index and non-alcoholic fatty liver disease fibrosis scores in metabolic (dysfunction) associated fatty liver disease according to body mass index: failure in the prediction of advanced fibrosis in lean and morbidly obese individuals. Eur J Gastroenterol Hepatol 2022;34(1):98–103. doi:10.1097/MEG.0000000000001946, PMID:32976186.

[20] Bedogni G, Bellentani S, Miglioli L, Masutti F, Passalacqua M, Castiglione A, *et al*. The Fatty Liver Index: a simple and accurate predictor of hepatic steatosis in the general population. BMC Gastroenterol 2006;6:33. doi:10.1186/1471-230X-6-33, PMID:17081293.

[21] Sterling RK, Lissen E, Clumeck N, Sola R, Correa MC, Montaner J, *et al*. Development of a simple noninvasive index to predict significant fibrosis in patients with HIV/HCV coinfection. Hepatology 2006;43(6):1317–1325. doi:10.1002/hep.21178, PMID:16729309.

[22] Boursier J, Hagström H, Ekstedt M, Moreau C, Bonacci M, Cure S, *et al*. Non-invasive tests accurately stratify patients with NAFLD based on their risk of liver-related events. J Hepatol 2022;76(5):1013–1020. doi:10.1016/j.jhep.2021.12.031, PMID:35063601.

[23] Eslam M, Sanyal AJ, George J, International Consensus Panel. MAFLD: A Consensus-Driven Proposed Nomenclature for Metabolic Associated Fatty Liver Disease. Gastroenterology 2020;158(7):1999–2014.e1. doi:10.1053/j.gastro.2019.11.312, PMID:32044314.

[24] Alberti KG, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, Donato KA, *et al*. Harmonizing the metabolic syndrome: a joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity. Circulation 2009;120(16):1640–1645. doi:10.1161/CIRCULATIONAHA.109.192644, PMID:19805654.

[25] Kanwal F, Shubrook JH, Adams LA, Pfotenhauer K, Wai-Sun Wong V, Wright E, *et al*. Clinical Care Pathway for the Risk Stratification and Management of Patients With Nonalcoholic Fatty Liver Disease. Gastroenterology 2021;161(5):1657–1669. doi:10.1053/j.gastro.2021.07.049, PMID:34602251.

[26] Younossi ZM, Corey KE, Alkhouri N, Noureddin M, Jacobson I, Lam B, *et al*. Clinical assessment for high-risk patients with non-alcoholic fatty liver disease in primary care and diabetology practices. Aliment Pharmacol Ther 2020;52(3):513–526. doi:10.1111/apt.15830, PMID:32598051.

[27] Huang JF, Tsai PC, Yeh ML, Huang CF, Huang CI, Hsieh MH, *et al*. Risk stratification of non-alcoholic fatty liver disease across body mass index in a community basis. J Formos Med Assoc 2020;119(1):89–96. doi:10.1016/j.jfma.2019.03.014, PMID:30952479.

[28] Lonardo A, Arab JP, Arrese M. Perspectives on Precision Medicine Approaches to NAFLD Diagnosis and Management. Adv Ther 2021;38(5):2130–2158. doi:10.1007/s12325-021-01690-1, PMID:33829368.

[29] Fracanzani AL, Petta S, Lombardi R, Pisano G, Russello M, Consonni D, *et al*. Liver and Cardiovascular Damage in Patients With Lean Nonalcoholic Fatty Liver Disease, and Association With Visceral Obesity. Clin Gastroenterol Hepatol 2017;15(10):1604–1611.e1. doi:10.1016/j.cgh.2017.04.045, PMID:28554682.

[30] Zou B, Yeo YH, Nguyen VH, Cheung R, Ingelsson E, Nguyen MH. Prevalence, characteristics and mortality outcomes of obese, nonobese and lean NAFLD in the United States, 1999-2016. J Intern Med 2020;288(1):139–151. doi:10.1111/joim.13069, PMID:32319718.

[31] Wei JL, Leung JC, Loong TC, Wong GL, Yeung DK, Chan RS, *et al*. Prevalence and Severity of Nonalcoholic Fatty Liver Disease in Non-Obese Patients: A Population Study Using Proton-Magnetic Resonance Spectroscopy. Am J Gastroenterol 2015;110(9):1306–1314. doi:10.1038/ajg.2015.235, PMID:26215532.

[32] Kim M, Yoon EL, Cho S, Lee CM, Kang BK, Park H, *et al*. Prevalence of advanced hepatic fibrosis and comorbidity in metabolic dysfunction-associated fatty liver disease in Korea. Liver Int 2022;42(7):1536–1544. doi:10.1111/liv.15259, PMID:35338555.
[33] Liu CJ. Prevalence and risk factors for non-alcoholic fatty liver disease in Asian people who are not obese. J Gastroenterol Hepatol 2012;27(10):1555–1560. doi:10.1111/j.1440-1746.2012.07222.x, PMID:22741595.
[34] Petersen KF, Dufour S, Feng J, Befroy D, Dziura J, Dalla Man C, *et al*. Increased prevalence of insulin resistance and nonalcoholic fatty liver disease in Asian-Indian men. Proc Natl Acad Sci U S A 2006;103(48):18273–18277. doi:10.1073/pnas.0608537103, PMID:17114290.
[35] Kaya E, Yilmaz Y. Epidemiology, natural history, and diagnosis of metabolic dysfunction-associated fatty liver disease: a comparative review with nonalcoholic fatty liver disease. Ther Adv Endocrinol Metab 2022;13:20420188221139650. doi:10.1177/20420188221139650, PMID:36533185.
[36] Younossi ZM, McCullough AJ, Ong JP, Barnes DS, Post A, Tavill A, *et al*. Obesity and non-alcoholic fatty liver disease in chronic hepatitis C. J Clin Gastroenterol 2004;38(8):705–709. doi:10.1097/01.mcg.0000135372.10846.2a, PMID:15319656.
[37] Adinolfi LE, Restivo L, Zampino R, Guerrera B, Lonardo A, Ruggiero L, *et al*. Chronic HCV infection is a risk of atherosclerosis. Role of HCV and HCV-related steatosis. Atherosclerosis 2012;221(2):496–502. doi:10.1016/j.atherosclerosis.2012.01.051, PMID:22385985.
[38] Li J, Yang HI, Yeh ML, Le MH, Le AK, Yeo YH, *et al*. Association Between Fatty Liver and Cirrhosis, Hepatocellular Carcinoma, and Hepatitis B Surface Antigen Seroclearance in Chronic Hepatitis B. J Infect Dis 2021;224(2):294–302. doi:10.1093/infdis/jiaa739, PMID:33249474.
[39] Joo EJ, Chang Y, Yeom JS, Ryu S. Hepatitis B virus infection and decreased risk of nonalcoholic fatty liver disease: A cohort study. Hepatology 2017;65(3):828–835. doi:10.1002/hep.28917, PMID:28035771.
[40] Fedchuk L, Nascimbeni F, Pais R, Charlotte F, Housset C, Ratziu V, LIDO Study Group. Performance and limitations of steatosis biomarkers in patients with nonalcoholic fatty liver disease. Aliment Pharmacol Ther 2014;40(10):1209–1222. doi:10.1111/apt.12963, PMID:25267215.
[41] Kaya E, Bakir A, Kani HT, Demirtas CO, Keklikkiran C, Yilmaz Y. Simple Noninvasive Scores Are Clinically Useful to Exclude, Not Predict, Advanced Fibrosis: A Study in Turkish Patients with Biopsy-Proven Nonalcoholic Fatty Liver Disease. Gut Liver 2020;14(4):486–491. doi:10.5009/gnl19173, PMID:31530739.
[42] Jafarov F, Kaya E, Bakir A, Eren F, Yilmaz Y. The diagnostic utility of fibrosis-4 or nonalcoholic fatty liver disease fibrosis score combined with liver stiffness measurement by fibroscan in assessment of advanced liver fibrosis: a biopsy-proven nonalcoholic fatty liver disease study. Eur J Gastroenterol Hepatol 2020;32(5):642–649. doi:10.1097/MEG.0000000000001573, PMID:31651653.