Lifestyle changes during the coronavirus disease 2019 pandemic impact metabolic dysfunction–associated fatty liver disease

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Citation	Liver International. 42(5); 995-1004.
Issue Date	2022-05
Version of Record	2022-03-18
Type	Journal Article
Textversion	Author
Highlights	◇新型コロナウイルス感染症（COVID-19）のパンデミック前後で健康診断受診者のデータを解析した結果、代謝異常関連脂肪肝（MAFLD）の患者数が増加していることが判明。 ◇MAFLD 患者増加には夜食・飲酒・欠食等が関連。
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Supporting Information	Supporting Information is available at https://doi.org/10.1111/liv.15158
DOI	10.1111/liv.15158

概要	研究グループは、健診受診者の COVID-19 パンデミック前後における臨床検査データや生活習慣に関するアンケート結果を解析し、COVID-19 パンデミック後に MAFLD 患者が増加したこと、また、MAFLD 患者増加には夜食・飲酒・欠食（1 日 2 食のみ)が関与することを明らかにしました。 ‘新型コロナウイルス感染症のパンデミックで脂肪肝が増加－背景に夜食や欠食などの生活習慣の変化’. 大阪市立大学. https://www.osaka-cu.ac.jp/ja/news/2021/220324 (参照 2022/0/03/24)

Article type : Original Article

Editor : Luca Valenti

Lifestyle changes during the coronavirus disease 2019 pandemic impact metabolic dysfunction–associated fatty liver disease

Short title: COVID-19 lifestyle changes affect MAFLD

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Word counts for main body (from title page to the beginning of the references): 3742

Total number of figure and tables: 6

List of abbreviations:

ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; BP, blood pressure; CI, confidence interval; COVID-19, coronavirus disease 2019; DM, diabetes mellitus; FIB-4, Fibrosis-4; GGT, gamma-glutamyl transferase, HbA1c, glycosylated hemoglobin A1c; HBsAg, hepatitis B surface antigen; HCV-Ab, anti-hepatitis C virus antibody; HDL-C, high-density lipoprotein-cholesterol; HR, hazard ratio; MAFLD, metabolic dysfunction–associated fatty liver disease; NAFLD, nonalcoholic fatty liver disease; TG, triglycerides; WC, waist circumference.

Disclosures/Conflicts of interest: All authors declare that they have no conflicts of interest to disclose.

Grant support:

This work was supported by a Grant-in-Aid for Scientific Research (C) from JSPS KAKENHI 17K09437 (to HF).

Writing assistance: Not applicable.

Author Contributions: Writing: HF; data collection: SF, TK, AN, YN, YN, ST, and TW; statistical analyses: HF and NN; revision and editing: HF, YK, and NK; acceptance of the final version: all authors.

Data transparency statement: Data are available on reasonable request to the corresponding author.

Acknowledgments:

The authors are grateful to Tetsuya Okawa for technical assistance. The authors also thank BioScienceWriters (https://www.biosciencewriters.com/) for English language editing.

Abstract

BACKGROUND & AIMS: The coronavirus disease 2019 (COVID-19) pandemic precipitated lifestyle changes. We aimed to clarify whether COVID-19–induced lifestyle changes affected the development of metabolic dysfunction–associated fatty liver disease (MAFLD).

METHODS: This retrospective longitudinal study included 973 participants who underwent health checkups between 2018 and 2020. We used data from the MedCity21 health examination registry. Participants' clinical characteristics and lifestyle habits were investigated. Independent lifestyle predictors of MAFLD development before the pandemic (2018–2019) and during the pandemic (2019–2020) were identified using logistic regression analysis.

RESULTS: In 2018, 261 (27%) patients were diagnosed with MAFLD. Before the pandemic, 22 patients developed new MAFLD. During this time, routine late-night meals were identified as an independent lifestyle predictor of MAFLD development (hazard ratio [HR] 2.54, 95% confidence interval [CI] 1.02–6.36, P=0.046). In contrast, 44 patients developed new MAFLD during the pandemic. During this time, higher daily alcohol intake was identified as an independent lifestyle predictor of MAFLD development (HR 1.03, 95% CI 1.01–1.05, P=0.008). In participants aged <60 years, daily alcohol intake and the proportion of participants who ate 2 times/day were significantly higher in patients who developed MAFLD during the pandemic than in those who did not. In participants aged ≥60 years, no lifestyle habits were associated with MAFLD development before or during the pandemic.

CONCLUSIONS: New MAFLD diagnoses increased during the COVID-19 pandemic. Changes in lifestyle factors, particularly in those aged <60 years, must be monitored and addressed as the pandemic continues.

Keywords: physical activity, body mass index, obesity, nonalcoholic fatty liver disease

Lay summary:

The impact of the lifestyle changes associated with the COVID-19 pandemic on metabolic dysfunction-associated fatty liver disease (MAFLD) remains unknown. Late-night meals before the coronavirus disease 2019 (COVID-19) pandemic and daily alcohol intake during the COVID-19 pandemic were extracted as independent lifestyle predictors of MAFLD development. These results are relevant for patient lifestyle counseling to prevent the increasing number of patients with MAFLD in the COVID-19 pandemic.

Introduction

The coronavirus disease 2019 (COVID-19) outbreak in December 2019 spread rapidly throughout the world and was declared a global pandemic by the World Health Organization in March 2020.[1] The Japanese government declared a state of emergency on April 7, 2020 because of a rapid increase in the number of patients affected by COVID-19. Under this declaration, nonessential businesses, schools, and recreational facilities were urged to close, while individuals were strongly encouraged to stay at home except for essential activities. The COVID-19 crisis forced lifestyle changes, the so-called "new normal." For example, many people now work from home and have little contact with people other than their family members. These changes potentially led to less physical activity, altered daily life rhythms, and unhealthy lifestyles.[2] They also may have resulted in increases in body weight and body mass index (BMI), potentially increasing the incidence of obesity and obesity-related disorders.[3, 4]

Because of widespread obesity, nonalcoholic fatty liver disease (NAFLD) is one of the leading causes of liver disease worldwide. The prevalence of NAFLD globally is currently estimated to be approximately 25%.[5] NAFLD comprises a broad spectrum of diseases ranging from nonalcoholic fatty liver to nonalcoholic steatohepatitis to cirrhosis and/or hepatocellular

carcinoma.[6, 7] In 2020, an international consensus panel suggested that NAFLD should be renamed metabolic dysfunction–associated fatty liver disease (MAFLD) and proposed new information regarding its diagnosis.[8, 9]

The impact of "new normal" lifestyles on the incidence of MAFLD is unknown. We hypothesized that new lifestyles resulting from the COVID-19 pandemic affect the development of MAFLD. Thus, we aimed to determine whether lifestyle changes associated with the COVID-19 pandemic increased the incidence of MAFLD and to identify specific lifestyle changes associated with the development of MAFLD.

Methods

Study design and population

This retrospective longitudinal study of MAFLD was based on an analysis of data obtained from the MedCity21 health examination registry, and was performed in Osaka, the second-largest city in Japan. The MedCity21 health examination registry protocol included regular comprehensive health assessments and was conducted in full accordance with the tenets of the Declaration of Helsinki (approval no. 2927).[10] The protocol for the current study was approved by the ethics committee of the Graduate School of Medicine, Osaka City University (approval no. 2021-027; April 26, 2021). Written informed consent was waived because of the study's retrospective nature. To allow participants the opportunity to refuse to participate, we provided a web-based opt-out option.

We selected all individuals who underwent abdominal ultrasonography in 2018 and analyzed those who returned for routine follow-up until 2020. Repeat visitors who underwent medical checkups during period 1 (July 1–December 31, 2018), period 2 (July 1–December 31, 2019), and period 3 (July 1–December 31, 2020) were included (Figure 1). We defined the

duration from period 1 to period 2 as the time before the COVID-19 pandemic and the duration from period 2 to period 3 as the time during the COVID-19 pandemic. The exclusion criteria were as follows: lack of data regarding hepatitis B surface antigen (HBsAg)/anti-hepatitis C antibody (HCV-Ab), serologic positivity for HBsAg and HCV-Ab, or any missing data components. Health checkup was postponed for examinees who were PCR positive for COVID-19 within 4 weeks from the date of examination or had symptoms (cough, fever, etc.) suspicious for COVID-19 infection on the date of examination. While analyzing lifestyle associations with the development of MAFLD, we also performed subgroup analyses according to age because lifestyle patterns differ between younger and older populations.[11, 12]

Clinical assessment

Data regarding the participants' clinical and demographic characteristics (age, sex, height, body weight, and blood pressure [BP]) and laboratory parameters were measured and recorded during all medical checkups. BMI was calculated as weight (kg) / height $(m)^2$. Waist circumference (WC) was measured with a non-stretchable tape at the level of the umbilicus with the participant in the standing position during the late phase of expiration. All data related to lifestyle habits (smoking history, physical exercise frequency, sleeping duration, meals per day, and late-night meals) and medical history were obtained using a questionnaire. A routine late-night meal was defined as having dinner within 2 hours before bedtime at least 3 times per week. The presence or absence of diabetes mellitus (DM), hypertension, and dyslipidemia was recorded; these diseases were diagnosed according to standard criteria.[8, 13-15] After an overnight fast, blood samples were collected and analyzed using standard laboratory procedures to obtain values for the complete blood count, aspartate aminotransferase (AST), alanine aminotransferase (ALT), albumin, gamma-glutamyl transferase (GGT), total cholesterol, low-density lipoprotein-cholesterol,

high-density lipoprotein-cholesterol (HDL-C), triglycerides (TG), fasting plasma glucose, and glycosylated hemoglobin A1c (HbA1c). Lumipulse HBsAg and HCV assays (Fujirebio Inc., Tokyo, Japan) were used to assess serologic markers, including HBsAg and anti-HCV-Ab. The Fibrosis-4 (FIB-4) index was calculated using available parameters.[16]

Fatty liver diagnosis

Fatty liver was diagnosed via abdominal ultrasonography using a Toshiba Aplio 500 Ultrasound Machine (Toshiba Medical Systems Corporation, Ohtawara, Japan). Abdominal ultrasonography was performed by experienced sonographers who were registered with the Japan Society of Ultrasonics in Medicine. Hepatic steatosis was semi-quantified according to the criteria described by Hamaguchi et al., which are based on the presence of hepatorenal echo contrast, a bright liver, deep attenuation, and vessel blurring.[17]

Alcohol intake screening and definition of alcoholic liver disease

Daily alcohol consumption was calculated in grams using our modified template.[18, 19] Frequency of alcohol intake was classified as 1 day per week, 3 days per week, or 7 days per week. Each participant's average alcohol consumption was classified as 10, 30, 50, or 70 g per day on days when alcohol was consumed. Daily alcohol consumption (g/day) was calculated as follows: (frequency of alcohol intake × average alcohol consumption [g]) / 7. Habitual alcohol intake was defined as alcohol intake of 1–59 g/day.

Diagnostic criteria and definition of MAFLD and NAFLD

MAFLD was defined as hepatic steatosis identified on abdominal ultrasonography and the presence of any 1 of these 3 conditions: overweight/obesity, DM, or evidence of metabolic

dysregulation. Metabolic dysregulation was defined as the presence of 2 or more of the following: WC ≥ 90 cm for men and ≥ 80 cm for women; BP ≥ 130/85 mmHg or current specific antihypertensive drug treatment; TG ≥ 150 mg/dL or current specific hypolipidemic drug treatment; HDL-C < 40 mg/dL for men and <50 mg/L for women; or prediabetes (fasting glucose 100–125 mmol/L or HbA1c 5.7%–6.4%).[8] We did not use the homeostasis model assessment of the insulin resistance score or high-sensitivity C-reactive protein level during this study. NAFLD was defined as ultrasonographic evidence of fatty liver disease and lack of excessive alcohol consumption (≥ 30 g/day for men and > 20 g/day for women).[6, 7] We defined "MAFLD development" as no MAFLD in the first year but the presence of MALD in the following year. "MAFLD resolution" was defined as the presence of MAFLD in the first year but no MALD in the following year.

Statistical analyses

Continuous variables were expressed as median (range), mean (standard deviation [SD]), or number (%). Categorical variables were expressed as frequency and percentage. Differences between groups were analyzed using the Wilcoxon rank-sum test for continuous variables and the Kruskal–Wallis test for categorical variables. Multiple comparisons were performed using Steel's multiple comparison test. A logistic regression model was used to identify independent predictors associated with MAFLD development.

Univariate (unadjusted) and multivariate (adjusted) hazard ratio (HR) estimates of the risk of MAFLD development were calculated to control for the effects of potential risk factors (confounders), including clinically important variables, such as age, sex, daily alcohol intake, BMI, ΔBMI, and ALT level. Data were expressed as HR and 95% confidence interval (CI). $P < 0.05$ was considered statistically significant. All statistical analyses were performed using JMP® 16.1.0 software (SAS Institute Inc., Cary, NC, USA).

Results

Participant characteristics

We enrolled 973 repeat visitors (487 females, 486 males) who underwent medical checkups during periods 1, 2, and 3 (Figure 1). Table 1 shows the participants' clinical characteristics. In period 1, the mean age was 52.5 years, mean BMI was 22.8 kg/m^2, and mean WC was 82.6 cm. In this period, 261 (27%) participants overall were diagnosed with MAFLD and 240 (25%) participants overall were diagnosed with NAFLD.

Table 2 shows the lifestyle habits of all participants during the 3 time periods. No significant changes in lifestyle habits were found before the COVID-19 pandemic (between periods 1 and 2) or during the pandemic (between periods 2 and 3).

Lifestyle habits related to MAFLD development

To determine the lifestyle characteristics of patients with newly developed or resolved MAFLD, we separately analyzed potential lifestyle habits associated with MAFLD development before and during the COVID-19 pandemic (Table 3). Before the pandemic, 22 patients developed new MAFLD from period 1 to period 2. Only the proportion of participants who consumed late-night meals was significantly higher among patients with newly developed MAFLD than among those without MAFLD (Table 3). During the pandemic, 44 patients developed new MAFLD from period 2 to period 3. Daily alcohol intake was significantly higher in patients with newly developed MAFLD than in those without MAFLD (mean 19.5 vs. 11.3 g/day, $P = 0.001$). Additionally, the proportions of current smokers and participants who ate 2 meals per day were significantly higher among patients with newly developed MAFLD than in those without MAFLD. Conversely, the proportions of never smokers and participants who ate 3 times per day were significantly lower

among patients with newly developed MAFLD than among those without MAFLD (Table 3). Regarding MAFLD resolution, there were no significant differences in lifestyle habits between patients with or without MAFLD resolution in both the time before the COVID-19 pandemic and the time during the COVID-19 pandemic (data not shown).

Relationships between clinical and laboratory data and MAFLD development

We evaluated associations between clinical and laboratory data and MAFLD development in the time before the COVID-19 pandemic and the time during the pandemic (Appendix Table 1). Both before and during the pandemic, patients who developed MAFLD had significantly higher levels of lipid parameters than those without MAFLD: TG before, 123 vs. 78.0 mg/dL ($P < 0.001$); TG during, 115 vs. 76.4 mg/dL ($P < 0.001$); LDL-C before, 129 vs. 110 mg/dL ($P = 0.003$); and LDL-C during, 123 vs. 110 mg/dL ($P = 0.002$). Patients with newly developed MAFLD also had significantly higher values for several other parameters both before and during the pandemic: ALT before, 23.2 vs. 17.5 U/L ($P = 0.003$); ALT during, 24.4 vs. 17.5 U/L ($P = 0.003$); fasting plasma glucose before, 105 vs. 99.3 mg/d ($P = 0.001$); fasting blood glucose during, 102 vs. 99.0 mg/dL ($P = 0.006$); BMI before, 24.0 vs. 21.5 kg/m^2 ($P < 0.001$); BMI during, 23.9 vs. 21.5 kg/m^2 ($P < 0.001$); body weight before, 66.6 vs. 57.6 kg ($P < 0.001$); body weight during, 65.6 vs. 57.6 kg ($P < 0.001$); WC before, 85.2 vs. 79.2 cm ($P < 0.001$); and WC during, 86.5 vs. 79.6 cm ($P < 0.001$) (Appendix Table 1).

Lifestyle habits as predictors associated with MAFLD development

Univariate and multivariate analyses were performed to explore lifestyle predictors of MAFLD development (Appendix Table 2 and Table 4). Univariate analyses showed that partaking in a routine late-night meal was a lifestyle predictor of MAFLD development in the time before the

COVID-19 pandemic (HR 2.47, 95% CI 1.05–5.82, $P = 0.038$). Lifestyle predictors of MAFLD development during the pandemic were higher daily alcohol intake (HR 1.02, 95% CI 1.01–1.04, $P = 0.004$), never smoking (HR 0.48, 95% CI 0.26–0.90, $P = 0.021$), current smoking (HR 2.34, 95% CI 1.08–5.08, $P = 0.031$), consuming 2 meals per day (HR 2.26, 95% CI 1.10–4.64, $P = 0.026$), and consuming 3 meals per day (HR 0.41, 95% CI 0.20–0.82, $P = 0.012$) (Appendix Table 2).

Multivariate adjusted HRs are presented in Table 4. In the time before the COVID-19 pandemic, partaking in a late-night meal remained an independent lifestyle predictor of MAFLD development (HR 2.54, 95% CI 1.02–6.36, $P = 0.046$), even after making adjustments for age, sex, BMI, ΔBMI, and ALT. During the pandemic, higher daily alcohol intake was the only independent lifestyle predictor of MALD development (HR 1.03, 95% CI 1.01–1.05, $P = 0.008$) after adjusting for the same factors (Table 4).

Lifestyle habits as predictors of MAFLD development considering interactive effects

We constructed a logistic regression model considering interactive effects between lifestyle factors (Appendix Table 3). In multivariate analysis, consuming a late-night meal was the only identified independent predictor of MAFLD development (HR 2.54, 95% CI 1.02–6.36, $P = 0.046$). There were no significant interactions between each factor and their respective differences.

Subgroup analysis according to age

Subgroup analysis of participants aged ≥ 60 years revealed no significant differences in lifestyle habits between patients with MALD development or without MALD development both before the pandemic and during the pandemic (Appendix Table 4). In participants < 60 years of age, there were also no significant differences in lifestyle habits between patients with or without MALD

development in the time before the COVID-19 pandemic. However, daily alcohol intake (mean 21.4 vs. 10.8 g/day, $P < 0.001$) and the proportion of participants who ate 2 times per day (28% vs 14%, $P = 0.029$) were significantly higher, and the proportion of participants who ate 3 times per day (69% vs 85.4%, $P = 0.016$) was significantly lower among participants who developed MAFLD during the COVID-19 pandemic in this younger age group (Table 5).

We also performed subgroup analysis of clinical factors in participants aged < 60 years (Appendix Table 5). Both before and during the pandemic, participants with newly developed MAFLD had significantly higher lipid parameters, ALT, fasting plasma glucose, BMI, body weight, and WC than participants without MAFLD. Moreover, during the COVID-19 pandemic, patients who developed new MAFLD had a significantly higher rate of hypertension, as well as higher age, diastolic BP, uric acid, AST, and GGT than those who did not develop MAFLD (Appendix Table 5).

Discussion

To the best of our knowledge, there have been no previous reports of increased MAFLD incidence associated with the lifestyle changes necessitated by the COVID-19 pandemic. In the current longitudinal study, we found that routine late-night meals before the COVID-19 pandemic and higher daily alcohol intake during the COVID-19 pandemic were independent lifestyle predictors of MAFLD development in a Japanese population.

Role of dietary habits in MAFLD development before and during the COVID-19 pandemic

Habitual late-night meals were identified as an independent risk factor for NAFLD in a previous Japanese questionnaire-based study.[20] Nishi et al. reported that participants who ate before bedtime had a higher risk of NAFLD than those who did not (adjusted odds ratio [OR] 2.15, 95% CI 1.03–4.46).[20] High energy intake at night might decrease insulin sensitivity,[21] which is strongly

associated with fatty liver development.[22] Additionally, recent evidence revealed that lifestyle changes post-COVID-19 were associated not only with obesity[3, 4] and obesity-related diseases (NAFLD[23]) but also with less healthy dietary habits.[12]

In this study, we confirmed routine late-night meals as a predictor of MAFLD development using multivariate analysis of interaction effects (Appendix Table 3). These results suggested that consuming late-night meals was the most important predictor of MAFLD development, regardless of the COVID-19 pandemic. Higher alcohol intake was identified as a new predictor of MAFLD development during the COVID-19 pandemic. A possible explanation is that participants with a predisposition towards late-night eating likely developed MAFLD by the second year of the study (2019), and increased alcohol intake during the COVID-19 pandemic may have masked the effects of late-night meals on MAFLD development during this later time period.

Role of alcohol intake in MAFLD development before and during the COVID-19 pandemic

We identified higher daily alcohol intake as an independent lifestyle predictor of MAFLD development during the COVID-19 pandemic mainly among participants aged < 60 years, which represents a major proportion of the working-age population. One report showed that medical students in Portugal reduced their alcohol intake during the pandemic lockdown.[24] Conversely, a questionnaire-based survey of approximately 10,000 Japanese adults revealed that during the stay-at-home request, the number of people reporting increased alcohol intake was higher in participants with extended at-home time than in those without.[25]

Because Alpers et al., who studied alcohol intake in the early stages of the COVID-19 pandemic, reported that people aged 30–39 years had a higher OR of increased drinking during lockdown than their oldest age group (OR 3.1, 95% CI 2.4–3.8),[27] we hypothesized that the impact of the COVID-19 pandemic on alcohol intake, as well as other lifestyle habits, would differ by

age.[11, 12] As expected, our results were consistent with those of Alpers et al.[26] One possible reason for increased alcohol consumption is the increase in teleworking from home. A report from Norway revealed that working from home for > 15 h per week was significantly associated with increased alcohol consumption (OR 1.67, 95% CI 1.30–2.16).[27] Therefore, these data suggest that the increase in teleworking during the COVID-19 pandemic led to an increase in alcohol consumption among the working-age population.

Other factors affecting MAFLD development before and during the COVID-19 pandemic

Current smoking and consuming 2 meals per day were identified as predictors of MAFLD development during the COVID-19 pandemic only on univariate analysis (Appendix Table 2). A recent 31-year cohort study demonstrated that individuals with persistent exposure to passive smoking between childhood and adulthood had the highest risk of fatty liver (relative risk 1.99, 95% CI 1.14–3.45).[28] Moreover, Alhussain et al. found that reduced meal frequency associated with irregular meal timing could cause weight gain and increase the levels of hunger-related hormones (glucagon-like peptide 1 and peptide YY), ultimately leading to metabolic disturbance.[29] Future large-scale prospective studies are necessary to clarify whether these lifestyle habits are indeed involved in MAFLD development.

Study strengths and weaknesses

The main strength of our study lies in its research design. We designed a longitudinal study to provide objective evidence regarding the effects of COVID-19–related lifestyle changes over time in the same individuals. Nevertheless, several limitations must be considered when interpreting our results. First, this was a retrospective, single-center study performed in Osaka, second largest city in Japan. Interestingly, recent study demonstrated that there are significant differences in the

proportion of Japanese NAFLD patients by region, but not by population-setting (urban or rural) or by per-person income data.[30] Second, selection bias was a major limitation. Our health checkup facility was temporarily closed from April 8, 2020 to May 31, 2020 to prevent the spread of COVID-19. Consequently, 30% of patients examined consecutively in 2018 and 2019 were not examined in 2020. This resulted in a smaller number of individuals who visited in all 3 periods and were thus included in this study. Third, we did not use the International Physical Activity Questionnaire, an internationally standardized questionnaire.[31] Thus, the assessment of physical activity levels may be insufficient to draw reliable conclusions about this potential predictive factor. Finally, most participants were healthy enough to engage in work and were sufficiently conscious of their health to voluntarily undergo health checkups.[32] Therefore, the study results may not apply to individuals who are less healthy.

In conclusion, we demonstrated that routinely partaking in late-night meals during the time before the COVID-19 pandemic and higher daily alcohol intake during the COVID-19 pandemic were independent lifestyle predictors for MAFLD development. These results highlight the need to avoid excessive alcohol intake as the pandemic continues to affect lifestyle patterns.

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Figure 1.

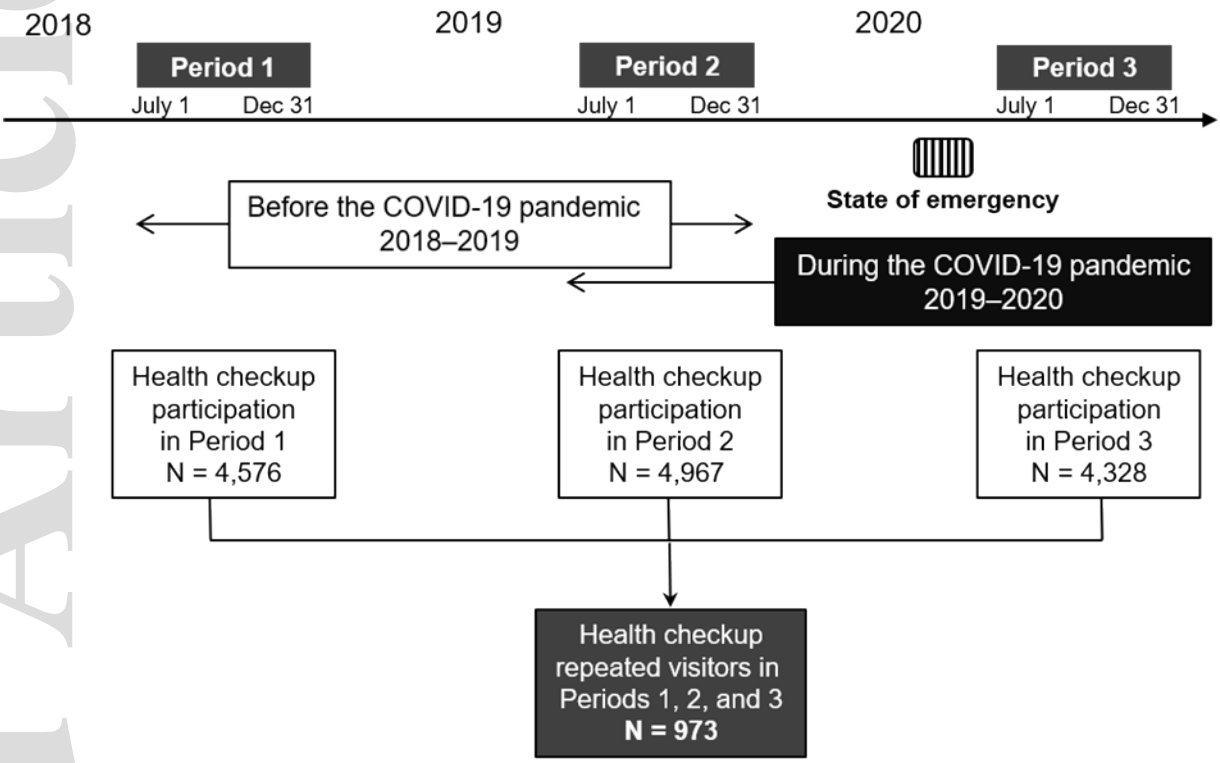

Figure legends

Figure 1. Study flow chart. COVID-19: coronavirus disease 2019.

Table 1. Clinical characteristics of participants (N=973)

Variables	Period 1	Period 2	Period 3
Age (y)†	52.5 (10.4)	53.6 (10.4)	54.6 (10.4)
Sex (female/male)‡	487/486	487/486	487/486
BMI (kg/m^2)†	22.8 (3.4)	22.8 (3.5)	22.9 (3.5)
WC (cm)†	82.6 (9.5)	83.3 (9.7)	83.6 (9.9)
MAFLD§	261 (27)	252 (26)	272 (28)
NAFLD§	240 (25)	229 (24)	246 (25)
Hypertension§	206 (21)	241 (25)	317 (33)
DM§	75 (8)	85 (9)	97 (10)
Dyslipidemia§	441 (45)	427 (44)	510 (52)
SBP (mmHg)†	118.3 (15.5)	119.8 (15.4)	121 (17.9)
DBP (mmHg)†	74.2 (10.7)	74.8 (10.6)	75.6 (12.0)
TC (mg/dL)†	198.8 (31.9)	197.9 (32.1)	204 (33.9)
TG (mg/dL)†	97.6 (77.6)	97.5 (74.8)	99.3 (66.8)
HDL-C (mg/dL)†	61.3 (15.9)	61.0 (15.8)	61.5 (16.1)
LDL-C (mg/dL)†	112.3 (28.0)	112.9 (27.9)	116.0 (29.0)
Non-HDL-C (mg/dL)†	137.5 (30.9)	136.8 (31.4)	142.8 (32.1)
Uric acid (mg/dL)†	5.3 (1.4)	5.3 (1.3)	5.3 (1.3)
AST (U/L)†	21.5 (7.6)	21.9 (8.3)	22.1 (7.9)
ALT (U/L)†	21.2 (13.5)	21.7 (14.0)	22.2 (14.3)
GGT (U/L)†	34.4 (41.2)	34.2 (36.8)	34.2 (37.7)
Albumin (g/dL)†	4.2 (0.3)	4.3 (0.3)	4.3 (0.3)
Platelet count (×10^9/L)†	229 (54.8)	230 (57.7)	239 (60.7)
Fasting plasma glucose (mg/dL)†	103 (14.4)	103 (15.6)	103 (15.2)
HbA1c (%)†	5.7 (0.5)	5.7 (0.5)	5.7 (0.5)
FIB-4 index score†	1.21 (0.57)	1.24 (0.60)	1.22 (0.59)

ALT: alanine aminotransferase; AST: aspartate aminotransferase; BMI: body mass index; DM: diabetes mellitus; DBP: diastolic blood pressure; FIB-4: Fibrosis-4; GGT: gamma-glutamyl transferase; HDL-C: high-density lipoprotein cholesterol; LDL-C: low-density lipoprotein cholesterol; MAFLD: metabolic dysfunction–associated fatty liver disease; NAFLD: non-alcoholic fatty liver disease; SBP: systolic blood pressure; TC: total cholesterol; TG: triglycerides; WC: waist circumference.

† Mean (standard deviation). ‡ Number. § Number (%).

Table 2. Lifestyle habits of participants (N=973)

Variables	Period 1	Period 2	Period 3	*P* value Period 1 vs. Period 2	*P* value Period 2 vs. Period 3
Alcohol intake (g/day) †	13.0 (17.7)	12.8 (17.2)	11.7 (17.0)	0.99	0.150
Smoking status ‡					
Never	572 (59)	572 (59)	567 (58)	0.99	1.00
Past	282 (29)	286 (29)	302 (31)		
Current	119 (12)	115 (12)	104 (11)		
Exercise (times/week) ‡					
< 1	547 (56)	510 (52)	495 (51)	0.21	0.30
1–2	275 (28)	302 (31)	278 (29)		
≥ 3	151 (16)	161 (17)	200 (20)		
Sleep duration (h/night) ‡					
< 6	360 (37)	356 (37)	328 (33)	0.99	0.28
6–8	583 (60)	597 (61)	621 (64)		
≥ 8	30 (3)	20 (2)	24 (3)		
Meals per day ‡					
1	4 (0.4)	5 (0.5)	5 (0.5)	0.38	0.60
2	118 (12)	135 (14)	122 (12.5)		
3	851 (87.6)	833 (85.5)	846 (87.0)		
Late-night meal ‡					
Yes	266 (27)	254 (26)	219 (23)	0.76	0.116
No	707 (73)	707 (74)	754 (77)		

† Mean (standard deviation). ‡ Number (%).

Table 3. Relationship between patient lifestyle habits and MAFLD development

	Before COVID-19 pandemic (2018–2019)			During COVID-19 pandemic (2019–2020)		
Variables[†]	**No (n=690)**	**Yes (n=22)**	***P* value**	**No (n=677)**	**Yes (n=44)**	***P* value**
Alcohol intake (g/day)[†]	12.0 (16.7)	15.8 (20.2)	0.30	**11.3 (16.1)**	**19.5 (19.1)**	**0.001**
Smoking status[‡]						
Never	433 (63)	14 (64)	1.00	**428 (63)**	**20 (45)**	**0.024**
Past	180 (26)	7 (32)	0.62	182 (27)	15 (34)	0.30
Current	77 (11)	1 (4)	0.50	**67 (10)**	**9 (21)**	**0.039**
Exercise (times/week)[‡]						
< 1	376 (54)	15 (68)	0.28	345 (51)	24 (55)	0.76
1–2	203 (29)	4 (18)	0.32	212 (31)	16 (36)	0.51
≥ 3	111 (17)	3 (14)	1.00	120 (18)	4 (9)	0.21
Sleep duration (h/night)[‡]						
< 6	259 (38)	7 (31)	0.66	254 (38)	15 (34)	0.75
6–8	407 (59)	14 (64)	0.83	406 (60)	28 (64)	0.75
≥ 8	24 (3)	1 (5)	0.55	17 (2)	1 (2)	1.00
Meals per day[‡]						
1	2 (0.3)	0 (0)	1.00	3 (0.4)	1 (2)	0.22
2	84 (12)	1(5)	0.50	**87 (13)**	**11 (25)**	**0.037**
3	604 (87.7)	21 (95)	0.50	**587 (86.6)**	**32 (73)**	**0.022**
Late-night meal[‡]						
Yes	**174 (25)**	**10 (45)**	**0.046**	177 (26)	10 (23)	0.86

COVID-19, coronavirus disease 2019; MAFLD: metabolic dysfunction–associated fatty liver disease.
[†] Mean (standard deviation). [‡] Number (%).
Note: Significant results are highlighted in bold.

Table 4. Results of multivariate analyses: Predictors of MAFLD development

	Before COVID-19 pandemic (2018–2019)		During COVID-19 pandemic (2019–2020)	
Variables	**Hazard ratio (95% CI)**	***P* value**	**Hazard ratio (95% CI)**	***P* value**
Age (y)	0.99 (0.94–1.03)	0.56	0.98 (0.95–1.02)	0.28
Sex (male)	1.08 (0.35–3.32)	0.90	1.17 (0.52–2.64)	0.71
BMI (kg/m^2)	**1.35 (1.15–1.58)**	**<0.001**	**1.38 (1.22–1.57)**	**<0.001**
ΔBMI (kg/m^2)	**2.07 (1.13–3.78)**	**0.013**	**2.94 (1.84–4.71)**	**<0.001**
ALT (U/L)	**1.05 (1.01–1.10)**	**0.025**	**1.03 (1.01–1.06)**	**0.013**
LIFESTYLE HABITS				
Alcohol intake (g/day)	1.01 (0.98–1.04)	0.38	**1.03 (1.01–1.05)**	**0.008**
Smoking status				
Never	4.59 (0.48–43.6)	0.185	0.77 (0.28–2.14)	0.61
Past	3.46 (0.37–32.7)	0.28	0.99 (0.36–2.71)	0.98
Current	N.A.		N.A.	
Meals per day				
1	N.A.	0.99	4.29 (0.25–73.5)	0.31
2	0.35 (0.04–2.93)	0.33	2.23 (0.95–5.22)	0.065
3	N.A.		N.A.	
Late-night meal †				
Yes	**2.54 (1.02–6.36)**	**0.046**	0.46 (0.20–1.07)	0.070

ALT: alanine aminotransferase; BMI: body mass index; CI: confidence interval; COVID-19: coronavirus disease 2019; MAFLD: metabolic dysfunction–associated fatty liver disease; N.A.: not applicable.

† Dinner within 2 hours before bedtime at least 3 times per week.

Note: Significant results are highlighted in bold.

Table 5. Relationship between patient lifestyle habits and MAFLD development in participants aged < 60 years (N=707)

	Before COVID-19 pandemic (2018–2019)			During COVID-19 pandemic (2019–2020)		
Variables[†]	**No (n=506)**	**Yes (n=18)**	***P* value**	**No (n=473)**	**Yes (n=36)**	***P* value**
Alcohol intake (g/day)[‡]	11.6 (16.4)	14.1 (20.2)	0.52	**10.8 (15.7)**	**21.4 (19.8)**	**<0.001**
Smoking status						
Never	324 (64)	13 (72)	0.48	305 (64)	18 (50)	0.105
Past	118 (23)	4 (22)	0.91	115 (24)	10 (28)	0.69
Current	64 (13)	1 (6)	0.77	53 (12)	8 (22)	0.061
Exercise (times/week)						
1	314 (62)	14 (79)	0.22	271 (57)	22 (61)	0.73
1–2	138 (27)	3 (17)	0.42	149 (32)	11 (31)	1.00
≥ 3	54 (11)	1 (4)	0.71	53 (11)	3 (8)	0.79
Sleep duration (h/night)						
< 6	204 (40)	7 (39)	1.00	189 (40)	13 (36)	0.73
6–8	290 (57)	11 (61)	0.81	278 (59)	22 (61)	0.86
≥ 8	12 (3)	0 (0)	1.00	6 (1)	1 (3)	0.40
Meals per day						
1	2 (1)	0 (0)	1.00	3 (0.6)	1 (3)	0.26
2	70 (13)	1 (6)	0.49	**65 (14)**	**10 (28)**	**0.029**
3	434 (86)	17 (94)	0.49	**405 (85.4)**	**25 (69)**	**0.016**
Late-night meal						
Yes	130 (26)	8 (44)	0.099	148 (31)	9 (25)	0.58

COVID-19: coronavirus disease 2019; MAFLD: metabolic dysfunction–associated fatty liver disease.

[†] Number (%). [‡] Mean (standard deviation).

Note: Significant results are highlighted in bold.