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Non-alcoholic fatty liver disease and sarcopenia additively increase mortality: a Korean nationwide survey

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Abstract

Background Sarcopenia is an independent risk factor not only for advanced-stage non-alcoholic fatty liver disease (NAFLD) but also for mortality. We investigated the association of sarcopenia and/or NAFLD with mortality among the Korean general population.

Methods Individuals aged 35–75 years without any history of cancer, ischaemic heart disease, ischaemic stroke, or secondary causes of chronic liver disease were selected from the Korean National Health and Nutrition Examination Surveys from 2008 to 2015. Their mortality data until December 2018 were retrieved from the National Death Registry. NAFLD and sarcopenia were defined by hepatic steatosis index and appendicular skeletal muscle mass divided by body mass index (BMI), respectively.

Results A total of 28 060 subjects were analysed [mean age, 50.6 (standard error, 0.1) years, 48.2 (0.3) % men]; the median follow-up duration was of 6.8 (interquartile range, 4.8, 8.4) years. NAFLD predicted mortality after adjustment for age, sex, BMI, hypertension, dyslipidaemia, and smoking (HR 1.32, 95% CI 1.03–1.70), but this prediction lost its statistical significance after additional adjustment for diabetes mellitus. In contrast, NAFLD with advanced fibrosis independently increased the risk of mortality after adjustment for all covariates (HR 1.68, 95% CI 1.02–2.79). Stratified analysis revealed that NAFLD and sarcopenia additively increased the risk of mortality as an ordinal scale (HR 1.46, 95% CI 1.18–1.81, P for trend = 0.001). The coexistence of NAFLD and sarcopenia increased the risk of mortality by almost twice as much, even after adjustment for advanced fibrosis (HR 2.18, 95% CI 1.38–3.44).

Conclusions Concurrent NAFLD and sarcopenia conferred a two-fold higher risk of mortality. The observation that NAFLD and sarcopenia additively increase mortality suggests that risk stratification would be helpful in predicting mortality among those with metabolic derangement.

Keywords Non-alcoholic fatty liver disease; Sarcopenia; Mortality; Nationwide survey

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Introduction

Non-alcoholic fatty liver disease (NAFLD) affects a quarter of the global population, and its prevalence continues to increase with age.[1,2] NAFLD, which is associated with a higher cardiovascular risk[3] and increased fibrosis leading to cirrhosis and hepatocellular carcinoma,[4] is an umbrella term that ranges from non-alcoholic fatty liver (NAFL) to non-alcoholic

steatohepatitis (NASH) and advanced fibrosis. In particular, advanced fibrosis is an independent risk factor for liver-related and non-liver-related mortality among subjects with NAFLD.[5–8]

The prevalence of sarcopenia is also rising as the global population ages,[9,10] and this significantly affects overall mortality not only in the elderly population but also in young adults.[11–13] Sarcopenia is also an independent risk factor for advanced-stage NAFLD (i.e. NASH and significant fibrosis) independent of obesity and insulin resistance.[14,15] Given that both NAFLD and sarcopenia prominently contribute to serious health consequences and share a common pathophysiology (e.g. insulin resistance and chronic inflammation),[16–18] it is of paramount importance to delineate the prognostic value of each disease entity using population-level data.

Here, we used a Korean nationwide survey and the National Death Registry to investigate the associations of NAFLD and sarcopenia with mortality to determine whether the presence of NAFLD and/or sarcopenia affects the mortality rate in the general population. We also assessed whether risk stratification based on subpopulation analysis could help clinicians predict prognosis in subjects with NAFLD and/or sarcopenia.

Methods

Study participants

The Korean Ministry of Health and Welfare designed the Korean National Health and Nutrition Examination Surveys (KNHANES), which has been conducted since 1998, to be representative of the Korean population using a stratified multistage probability sampling method; the survey has been described in detail elsewhere.[19] Briefly, the KNHANES recruit participants using a stratified multistage probability-based sampling design, and sampling weights are assigned to each respondent to ensure that the results are representative of the whole Korean population. The use of the KNHANES data was approved by the Institutional Review Board (IRB) of the Korea Centers for Disease Control and Prevention (IRB No. 2008-04EXP-01-C, 2009-01CON-03-2C, 2010-02CON-21-C, 2011-02CON-06-C, 2012-01EXP-01-2C, 2013-07CON-03-4C, 2013-12EXP-03-5C). IRB approval was not required for the use of KNHANES data for 2015 under the Bioethics Act. The study was conducted following the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments.

Individuals, 35 to 75 years of age who were enrolled in the KNHANES from January 2008 to December 2015 were eligible for this study. The exclusion criteria were as follows: alcohol consumption $>$210 g/week for men and $>$140 g/week for women[20]; positive serological markers for hepatitis B or C virus; history of cancer; history of ischaemic heart disease; and/or history of stroke. After this filtering, a total of 28 060 subjects were included in this study.

Definition of outcomes

Korean National Health and Nutrition Examination Surveys data were matched with those in the National Death Registry from the Korea National Statistical Office. The specific cause of death according to the International Classification of Diseases-10 (ICD-10) and date of death of the participants of KNHANES were available up to December 2018. Cause-specific mortality was identified by the following ICD-10 codes: cancer-specific mortality, C00–D48; cardiovascular disease (CVD)-specific mortality, I00–I99.

Assessment of metabolic parameters

Blood samples were drawn from the antecubital vein in the morning after subjects had fasted for at least 8 h. Samples were properly processed, immediately refrigerated at 2 to 8°C, and sent to a central laboratory. Fasting glucose, lipid profile (total cholesterol, high- and low-density lipoprotein cholesterol, and triglycerides), serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), gamma-glutamyl transferase, and creatinine were measured enzymatically (Hitachi Automatic Analyser 7600, Hitachi, Japan). Glycated haemoglobin was measured by high-performance liquid chromatography (HLC-723G7; Tosoh, Japan). Vitamin D was measured using a radioimmunoassay (1470 WIZARD gamma-Counter; PerkinElmer, Finland). Complete blood count was measured using an XE-2100D (Sysmex, Japan). Hepatitis B antigen was detected using an electrochemiluminescence immune assay (E-170; Roche, Germany), and hepatitis C antibody was detected using a chemiluminescent microparticle immunoassay (ARCHITECT i4000Sr; ABBOTT, Germany).

Diabetes mellitus (DM) was defined as a fasting plasma glucose concentration $\geq$7.0 mmol/L (126 mg/dL) or glycated haemoglobin $\geq$48 mmol/mol (6.5%), or reported use of anti-diabetic medication including insulin.[21] Hypertension was defined as a systolic blood pressure $\geq$140 mmHg, diastolic blood pressure $\geq$90 mmHg, or reported use of antihypertensive medication. Dyslipidaemia was defined as non-high-density lipoprotein cholesterol $\geq$190 mg/dL or reported use of lipid-lowering medication. Obesity was defined as a BMI $\geq$ 25 kg/m^2 based on the World Health Organization (WHO) Asia–Pacific criteria and the Korean Society for the Study of Obesity guideline.[22,23] Central obesity was defined as waist circumference $\geq$90 cm in men and $\geq$85 cm in women. Metabolic syndrome was diagnosed if $\geq$3 of the criteria were met, according to the National Cholesterol Education Program (NCEP) Adult Treatment panel (ATP) III

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revised criteria, with central obesity defined as above.[24] Chronic kidney disease was defined as an estimated glomerular filtration rate (eGFR) $<$ 60 mL/min/1.73 m^2, as calculated by the Modification of Diet in Renal Disease (MDRD) Study equation.[25] Vitamin D deficiency was defined as serum 25-hydroxyvitamin D $<$ 20 ng/ml.

Assessment of non-alcoholic fatty liver disease, fibrosis, and sarcopenia

Non-alcoholic fatty liver disease was defined as a hepatic steatosis index (HSI) $>$ 36 using the previously validated prediction model calculated as: 8 × ALT [U/L]/AST [U/L] + body mass index (BMI) [+2, if DM; +2 if female].[26] Liver fibrosis was defined as a fibrosis-4 index (FIB-4) $\geq$ 1.3 for age $\leq$65 and FIB-4 $\geq$ 2.0 for age $>$65; this was calculated as follows: age (years) × AST [U/L]/(platelet [10^9/L] × (ALT [U/L])$^{1/2}$.[27,28]

Appendicular skeletal muscle mass (ASM) was calculated as the sum of the lean mass in both arms and legs, which was assessed by dual energy X-ray absorptiometry (DXA; QDR Discovery; Hologic, Inc., Bedford, MA).[29] Sarcopenia was defined by the Foundation for the National Institutes of Health criteria as: ASM divided by BMI $\leq$ 0.789 for men and $\leq$0.512 for women.[30] DXA was performed between 2008 and 2011; thus, sarcopenia could be evaluated in 11 005 subjects.

Statistical analysis

The data are expressed as the mean (standard error, SE) or prevalence (SE) (%). Stratification variables and sampling weights were used as designated in KNHANES. A linear regression or logistic linear regression model was used to compare the clinical variables according to NAFLD, sarcopenia, or survival status. The Cox proportional hazards model was used to determine whether the presence of NAFLD, advanced fibrosis, or sarcopenia was independently associated with mortality. For sensitivity analysis, participants were categorized into four subgroups based on the presence of NAFLD or sarcopenia (Model 1, adjusted for age, sex, and BMI; Model 2, further adjusted for hypertension, dyslipidaemia, and smoking; Model 3, further adjusted for DM, chronic kidney disease, vitamin D status, and dyslipidaemia medication; and Model 4, further adjusted for advanced fibrosis). Hazard ratios (HRs) are presented with corresponding 95% confidence intervals (CIs) and *P*-values. *P*-values for trends were calculated assuming that NAFLD and sarcopenia corresponded to 1 point each as an ordinal scale. All statistical analyses were performed using a complex sample design and applying SPSS version 25.0 (IBM, Armonk, NY). $P < 0.05$ was considered statistically significant.

Results

Baseline characteristics of study subjects according to non-alcoholic fatty liver disease and advanced fibrosis status

A total of 28 060 subjects were included in the analysis; among them, 24.1 (SE, 0.3) % (unweighted *N* = 6488) were classified as NAFLD (*Table* 1). The mean age of the study population was 50.6 (SE, 0.1) years, and 48.2 (SE, 0.3) % were male.

Subjects with NAFLD had a higher prevalence of DM, hypertension, dyslipidaemia, obesity, and sarcopenia (age- and sex-adjusted $P < 0.001$ in all; *Table* 1). Their AST and ALT levels and eGFR were also worse than subjects without NAFLD (age- and sex-adjusted $P < 0.001$ in all; *Table* 1). Among subjects with NAFLD, those with advanced fibrosis were older and more obese and had higher AST, ALT, and GGT levels than those without advanced fibrosis (Supporting Information, *Table* S1).

Comparison of clinical characteristics according to survival status

Overall mortality was observed in 2.5 (SE, 0.1) % (unweighted *N* = 939) of all study subjects during the median follow-up period of 6.8 (interquartile range [IQR], 4.8, 8.4) years. Cancer-related mortality and CVD-related mortality were reported to be 0.9 (SE, 0.1) % and 0.5 (SE, 0.0) %, respectively. Deceased subjects were predominantly male (64.1 [SE, 2.0] % vs. 47.8 [SE, 0.3] %) and significantly older (mean age: 62.1 [SE, 0.5] years, vs. 50.3 [SE, 0.1] years) compared with survivors (Supporting Information, *Table* S2). After adjustment for age and sex, DM, dyslipidaemia, CKD, and smokers were more frequently found in the deceased subjects compared to survivors, whereas the prevalence of obesity was significantly lower in the deceased subjects ($P < 0.001$; Supporting Information, *Table* S2). The prevalence of sarcopenia was higher in the deceased subjects compared with survivors (21.0 [SE, 2.2] % vs. 8.6 [SE, 0.4] %; $P < 0.001$ in crude analysis), but this did not retain significance after adjustment for age and sex ($P = 0.087$; Supporting Information, *Table* S2).

Risk of mortality according to non-alcoholic fatty liver disease, advanced fibrosis, and sarcopenic status

Non-alcoholic fatty liver disease increased the risk of mortality by 42% in the age-, sex-, and BMI-adjusted model (HR, 1.42; 95% CI, 1.11–1.82; Model 1 in *Table* 2) and maintained its statistical significance after adjustment for hypertension,

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Table 1 Baseline clinical characteristics according to non-alcoholic fatty liver disease

	Total	NAFLD (−)	NAFLD (+)	*P* value*	*P* value**
Unweighted, *N*	28 060	21 572	6488		
Age (years)	50.6 (0.1)	50.7 (0.1)	50.1 (0.2)	<0.001	-
Gender (men, %)	48.2 (0.3)	46.9 (0.3)	52.1 (0.7)	<0.001	-
BMI (kg/m^2)	24.0 (0.0)	22.9 (0.0)	27.6 (0.0)	<0.001	<0.001
Waist circumference (cm)[a]	82.1 (0.1)	79.2 (0.1)	91.2 (0.1)	<0.001	<0.001
HbA1c (%)[a]	5.89 (0.01)	5.74 (0.01)	6.34 (0.02)	<0.001	<0.001
Total cholesterol (mg/dL)	193.5 (0.3)	191.2 (0.3)	200.6 (0.6)	<0.001	<0.001
Triglycerides (mg/dL)[a]	144.3 (0.9)	130.3 (0.9)	188.4 (2.1)	<0.001	<0.001
HDL-C (mg/dL)[a]	49.0 (0.1)	50.4 (0.1)	44.7 (0.2)	<0.001	<0.001
AST (U/L)[a]	22.4 (0.1)	21.3 (0.1)	25.6 (0.2)	<0.001	<0.001
ALT (U/L)[a]	22.2 (0.1)	18.1 (0.1)	35.2 (0.4)	<0.001	<0.001
GGT (U/L)[a]	35.7 (0.7)	32.1 (0.8)	47.5 (1.3)	<0.001	<0.001
Platelets (10^9/L)	255.4 (0.4)	253.5 (0.5)	261.4 (0.9)	<0.001	<0.001
eGFR (mL/min/1.73 m^2)[a]	92.3 (0.2)	92.6 (0.2)	91.7 (0.3)	0.001	<0.001
SBP (mmHg)	119.0 (0.2)	117.5 (0.2)	123.5 (0.2)	<0.001	<0.001
DBP (mmHg)	77.5 (0.1)	76.4 (0.1)	81.0 (0.2)	<0.001	<0.001
Diabetes mellitus (%)	11.5 (0.2)	6.6 (0.2)	26.8 (0.7)	<0.001	<0.001
Hypertension (%)	29.8 (0.4)	25.5 (0.4)	43.3 (0.8)	<0.001	<0.001
Dyslipidaemia (%)	15.4 (0.3)	12.4 (0.3)	24.9 (0.6)	<0.001	<0.001
Obesity (%)	34.8 (0.4)	19.0 (0.3)	84.7 (0.5)	<0.001	<0.001
Sarcopenia (%)	9.1 (0.4)	7.0 (0.4)	15.7 (1.0)	<0.001	<0.001
Chronic kidney disease (%)	1.8 (0.1)	1.7 (0.1)	2.2 (0.2)	0.012	<0.001
Vitamin D deficiency (%)	66.1 (0.7)	65.3 (0.7)	68.7 (1.0)	<0.001	<0.001
Smoking (%)					
Never	73.0 (0.3)	74.0 (0.4)	69.9 (0.7)	<0.001	0.262
Past	6.5 (0.2)	6.4 (0.2)	6.8 (0.4)		
Active	20.5 (0.;3)	19.6 (0.3)	23.3 (0.6)		

ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; DBP, diastolic blood pressure; eGFR, estimated glomerular filtration rate; GGT, gamma-glutamyl transferase; HDL-C, high-density lipoprotein cholesterol; NAFLD, non-alcoholic fatty liver disease; SBP, systolic blood pressure.
Values are presented as mean or % (standard error).
[a]Presented as median values (standard error).
*Without adjustment.
**With adjustment for age and sex.

Table 2 Hazard ratios for mortality according to non-alcoholic fatty liver disease, advanced fibrosis, and sarcopenia

	NAFLD		NAFLD with advanced fibrosis		Sarcopenia	
	HR (95% CI)	*P* value	HR (95% CI)	*P* value	HR (95% CI)	*P* value
Model 1	1.42 (1.11–1.82)	0.005	2.11 (1.30–3.43)	0.003	1.56 (1.17–2.08)	0.002
Model 2	1.32 (1.03–1.70)	0.027	2.10 (1.29–3.41)	0.003	1.53 (1.15–2.04)	0.004
Model 3	1.10 (0.85–1.42)	0.474	1.68 (1.02–2.76)	0.040	1.48 (1.11–1.98)	0.008

Hazard ratio for 6.8-year mortality was evaluated using multivariate Cox analysis.
Model 1: Adjusted for age, sex, and BMI.
Model 2: Adjusted for hypertension, dyslipidaemia, and smoking status in addition to the adjustments listed for Model 1.
Model 3: Adjusted for diabetes mellitus in addition to the adjustments listed for Model 2.

dyslipidaemia, and smoking status (HR 1.32, 95% CI, 1.03–1.70, Model 2 in *Table* 2). However, after additional adjustment for DM, NAFLD was not associated with overall mortality (Model 3 in *Table* 2; *Figure* 1A). In contrast, NAFLD with advanced fibrosis significantly increased overall mortality in the fully adjusted model (HR 1.68, 95% CI, 1.02–2.76; Model 3 in *Table* 2). Sarcopenia was also an independent risk factor for overall mortality in the fully adjusted model (HR 1.48, 95% CI, 1.11–1.98, Model 3 in *Table* 2).

Considering that sarcopenia was more prevalent in subjects with NAFLD (15.7% vs. 7.0%; *Table* 1), we stratified the study population into four subgroups by NAFLD and sarcopenia status to evaluate whether sarcopenia affects mortality in association with NAFLD. Compared with subjects with NAFLD but without sarcopenia, subjects with NAFLD and sarcopenia were older and had higher BMI (Supporting Information, *Table* S3). Stratified analysis according to sarcopenia and NAFLD confirmed that NAFLD without sarcopenia increased the risk of overall mortality significantly in the age-, sex-, and BMI-adjusted model compared with the reference group (those without NAFLD or sarcopenia; HR, 1.86, 95% CI, 1.27–2.74; Model 1 in Table 3). The statistical significance of this increase was maintained upon further adjustment for hypertension, dyslipidaemia, DM, chronic kidney disease, smoking, vitamin D, and advanced fibrosis (HR, 1.52, 95% CI, 1.02–2.25; Model 4 in Table 3). The coexistence of

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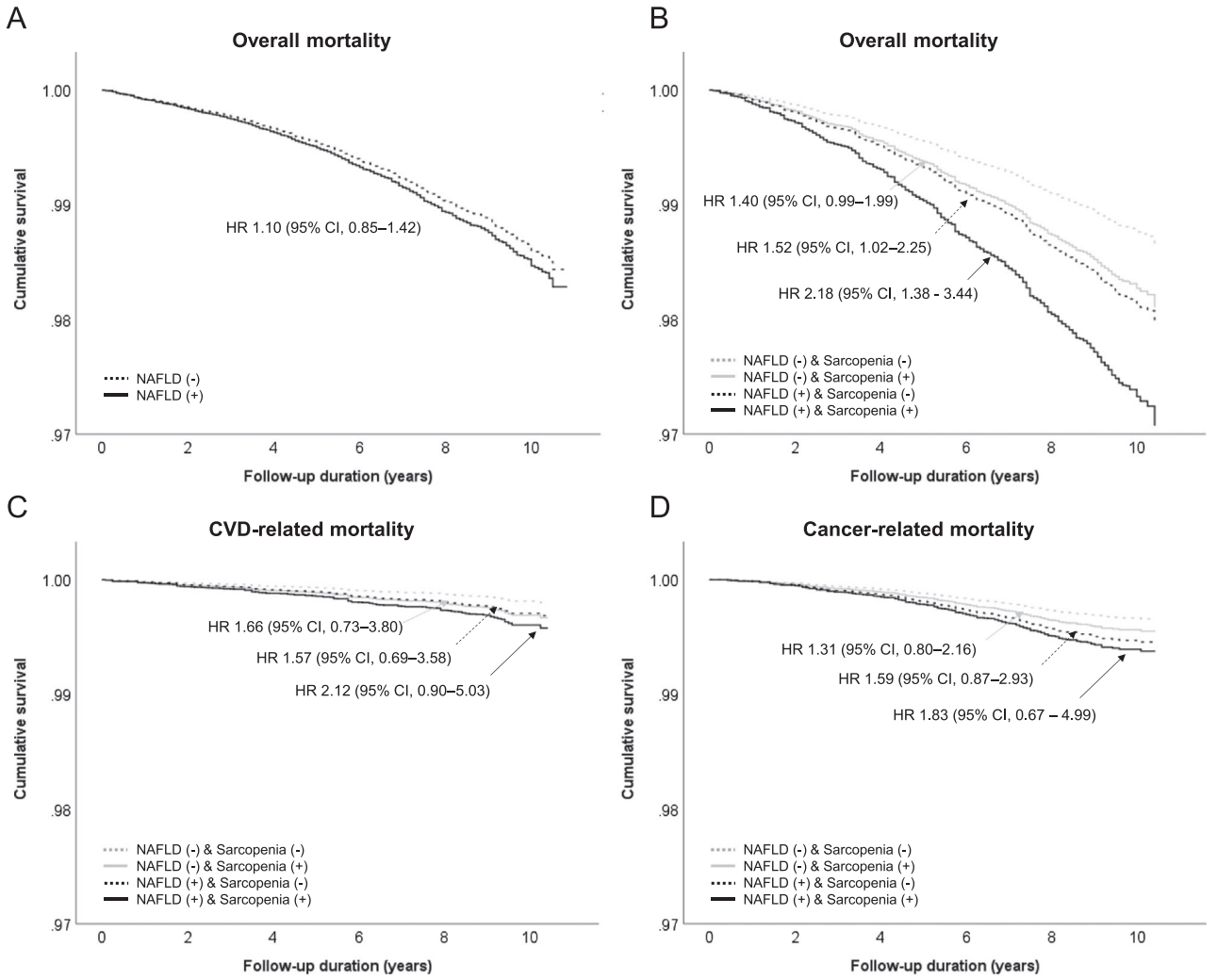

Figure 1 Overall and cause-specific mortality by NAFLD and sarcopenic status. Cumulative survivals according to (A) overall mortality in subjects with (solid black line) and without (dashed black line) NAFLD; and (B) overall, (C) CVD-related, and (D) cancer-related mortality by the presence of either NAFLD and/or sarcopenia for median 6.8 years of follow-up were analysed using Cox proportional hazards analysis. (1) No NAFLD and no sarcopenia (dashed grey line), (2) no NAFLD but sarcopenia (solid grey line), (3) NAFLD without sarcopenia (dashed black line), and (4) NAFLD with sarcopenia (solid black line) (unweighted *N* = 28 060; overall mortality, 2.5% (SE, 0.1%); CVD-related mortality, 0.5% (SE, 0.0%); cancer-related mortality, 0.9% (SE, 0.1%)). Hazard ratios (95% confidence intervals) were calculated after adjustment for age, sex, BMI, hypertension, dyslipidemia, smoking, and diabetes mellitus for *Figure* 1A (Model 3 in *Table* 2) or further adjustment for chronic kidney disease, vitamin D, dyslipidaemia medication, and advanced fibrosis for *Figure* 1B–D (Model 4 in *Table* 3 or Supporting Information, *Table* S5).

Table 3 Hazard ratios for mortality according to non-alcoholic fatty liver disease and sarcopenia

	NAFLD(−) Sarcopenia(−)	NAFLD(−) Sarcopenia(+)			NAFLD(+) Sarcopenia(−)			NAFLD(+) Sarcopenia(+)			
	HR	HR	95% CI	*P* value	HR	95% CI	*P* value	HR	95% CI	*P* value	*P* for trends
Model 1	1.00 (ref.)	1.53	1.09–2.15	0.013	1.86	1.27–2.74	0.002	2.85	1.83–4.44	<0.001	<0.001
Model 2	1.00 (ref.)	1.50	1.07–2.11	0.019	1.73	1.18–2.55	0.006	2.68	1.73–4.17	<0.001	<0.001
Model 3	1.00 (ref.)	1.39	0.98–1.98	0.065	1.45	0.98–2.14	0.062	2.08	1.32–3.27	0.002	0.001
Model 4	1.00 (ref.)	1.40	0.99–1.99	0.059	1.52	1.02–2.25	0.038	2.18	1.38–3.44	0.001	0.001

Hazard ratio for 6.8-year mortality was evaluated using multivariate Cox regression analysis.
Model 1: Adjusted for age, sex, and BMI.
Model 2: Adjusted for hypertension, dyslipidaemia, and smoking status in addition to the adjustments listed for Model 1
Model 3: Adjusted for diabetes mellitus, chronic kidney disease, vitamin D status, and dyslipidaemia medication in addition to the adjustments listed for Model 2.
Model 4: Adjusted for advanced fibrosis in addition to the adjustments listed for Model 3.

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NAFLD and sarcopenia approximately doubled the risk of overall mortality (HR, 2.18, 95% CI, 1.38–3.44 Model 4 in Table 3; *Figure* 1B). In this model, advanced fibrosis was also an independent risk factor for overall mortality (HR, 1.44, 95% CI, 1.13–1.84, Supporting Information, *Table* S4). Moreover, NAFLD and sarcopenia additively increased the risk of mortality as an ordinal scale (HR 1.46, 95% CI 1.18–1.81, *P* for trend = 0.001). However, neither CVD-related mortality nor cancer-related mortality were significantly increased in subjects with NAFLD and sarcopenia (Supporting Information, *Table* S5; *Figure* 1C,D).

Discussion

Non-alcoholic fatty liver disease and sarcopenia adversely affect metabolic health outcomes and pose an increasing global health and economic burden.[2,31] Therefore, in the current study, we assessed the prognostic values of NAFLD and sarcopenia in predicting mortality among the general population using a Korean nationwide survey and the National Death Registry. Using HSI $\geq$ 36 to define hepatic steatosis, we diagnosed a quarter of study subjects with NAFLD. We found that both sarcopenia and advanced fibrosis independently predicted mortality during the follow-up of 6.8 years, whereas NAFLD alone did not. The coexistence of NAFLD and sarcopenia was associated with an approximately 2.2-fold higher risk of mortality relative to the reference group without these conditions, and each condition additively contributed to an increased mortality.

Sarcopenia and NAFLD share common pathophysiological mechanisms, including insulin resistance and chronic inflammation.[32] Insulin resistance induces lipolysis in the adipose tissue and is associated with a higher circulating free fatty acid level,[33] which eventually leads to ectopic fat deposition in the muscle tissue (i.e. myosteatosis) and liver (i.e. simple steatosis). Insulin resistance promotes both *de novo* lipogenesis and gluconeogenesis in the liver through transcriptional activation of SREBP-1c and ChREBP,[34] and the subsequent increases in glucose flux and lipotoxicity exacerbate proteolysis in muscle.[32,35] Pro-inflammatory conditions associated with insulin resistance may directly stimulate protein catabolism, leading to a loss of muscle mass.[36] Myokines,[37] hepatokines,[38] and pro-inflammatory cytokines[36] have been suggested to contribute to the interplay between muscle and liver.[37,38] It thus appears that liver and muscle interact with each other to exacerbate the progression of both NAFLD and sarcopenia, although the underlying mechanisms and biological sequences responsible for such phenotypic changes need to be further elucidated. A more severe inflammatory response and/or insulin resistance may be driven by the coexistence of NAFLD and sarcopenia via changes in the interplay between liver and muscle, which might explain why the prevalence of sarcopenia exhibits a stepwise increase with increasing severity of NAFLD.[14,20,39] Furthermore, improvement in skeletal muscle mass during a 7-year follow-up period was associated with a decrease in the incidence of NAFLD,[40] which suggests that sarcopenia may be a predictor of NAFLD progression as well as a risk factor for NAFLD development.

Previous studies have yielded inconsistent results regarding the effect of NAFLD on mortality: NAFLD predicted mortality in some studies[41,42] but failed to do so in others.[43] Rather, advanced fibrosis was reported to be an independent risk factor for mortality among NAFLD subjects.[5,6] In the current study, stratified analysis according to the presence of NAFLD and/or sarcopenia indicated that subjects with NAFLD but without sarcopenia had a significantly increased risk of mortality compared with the reference group without NAFLD or sarcopenia, independent of advanced fibrosis. In the no-NAFLD population, the proportion of sarcopenic subjects was 7.0%; these individuals were significantly older and had a higher mortality rate compared with the reference group, which might attenuate the effect of NAFLD on mortality. Considering the conflicting data regarding the effect of NAFLD on mortality,[41–43] it is important to define the healthy reference group properly, and careful evaluation of metabolic and other risk factors should be made for the analysis. The apparent dose–response relationship between the risk of mortality and the presence of sarcopenia and/or NAFLD likely reflects that sarcopenia might increase the risk of mortality irrespective of the presence of NAFLD and vice versa.

In the current study, the coexistence of NAFLD and sarcopenia did not predict CVD- or cancer-related mortality although it significantly increased the overall mortality, which might be from the relatively small number of events studied in the present work. Recently, Golabi *et al*. reported sarcopenia independently contributed cardiac and cancer-specific mortality as well as overall mortality in the individuals with NAFLD using the US NHANES data.[44] In that study, 587 mortalities out of 4611 subjects were observed for 13.5 years; their long duration of follow-up might disclose the effect of sarcopenia with NAFLD more clearly. However, they did not exclude subjects with cancer or CVD at the baseline, although sarcopenia is closely associated with the risk of cancer and CVD.[45,46] The current study excluded subjects with previous history of cancer or CVD and adjusted for advanced fibrosis and BMI, well-known risk factors for mortality, which made it robust to demonstrate the predictive value of NAFLD and sarcopenia for mortality. Welch *et al*. also demonstrated that decreased initial muscle mass and the extent of the reduction in muscle mass measured by consecutive computed tomography scans are associated with increased risk of mortality in 83 patients with biopsy-proven cirrhosis.[47] These studies implicate that sarcopenia should be evaluated for risk stratification of mortality in subjects with the full spectrum of NAFLD. Further studies are needed to investigate whether the interplay between NAFLD and sarcopenia may exacerbate

each and/or additively increase the risk of cancer or CVD in the diverse populations.

Interestingly, obesity played a protective role in the mortality in the currently study. The J-shaped relationship between BMI and mortality has been reported across the diverse population.[48] In the Korean population, the lowest mortality rate was observed in obese subjects (25–29.9 kg/m^2),[49,50] which suggests that among the Korean population with BMI $<$ 30 kg/m^2, there might be an inverse association between their BMI and mortality rate. In addition, Asians have a relatively higher proportion of non-obese NAFLD, which exhibits distinct metabolic features and genetic alterations in *PNPLA3* and *TM6SF2* compared with obese NAFLD in Western population.[51–53] A recent study from the U.S. NHANES reported a higher mortality rate in subjects with non-obese NAFLD. Therefore, NAFLD subjects with a low to normal BMI should be carefully assessed for their long-term prognosis, although they may present with favourable metabolic profiles.[54]

A strength of our study is that we used nationwide survey data representing the Korean general population, and mortality data were retrieved from the National Death Registry from the Statistics Office of Korea. ICD-10 codes were used to define the cause of death. This study therefore has the unique advantages of utilizing survey data organized by the Korean government, and combining analysis of cross-sectional surveys (KNHANES) and the death registry to enable longitudinal outcome research. Our observation that the adverse joint effect of NAFLD and sarcopenia on mortality remained markedly significant even after adjustment for other metabolic comorbidities and advanced fibrosis emphasizes the need to perform stratified analysis by risk factors when seeking to predict long-term prognoses in subjects with metabolic derangements.

The current study has some limitations. First, we alternatively used the HSI and FIB-4 to define hepatic steatosis and advanced fibrosis, although histological or radiological assessment might be more accurate for confirming NAFLD. However, a number of studies on the diagnostic and prognostic performance of the HSI and FIB-4 indicated that they are acceptable surrogates for use in epidemiological studies.[26,28,55–57] Second, the follow-up duration varied among study participants due to the inherent nature in the study design, where ongoing annual surveys were sequentially incorporated into the current study with a single endpoint. Third, we could not analyse liver-related mortality in the current study. The KNHANES did not allow us to obtain or analyse data on liver-related mortality due to the small number of liver-related deaths. Finally, sarcopenia was not evaluable in all subjects, as DXA has not been performed since 2012. However, we could draw a relatively clear conclusion from this portion of the analysis.

With these caveats in mind, we herein report that concurrent NAFLD and sarcopenia additively increased the risk of mortality in the Korean general population. Our results are relevant to clinical practitioners because they may help guide the risk stratification of patients with NAFLD. Our finding that NAFLD increased the risk of mortality when stratified by sarcopenic status conveys an important message to clinicians, and may facilitate the prediction of clinical outcomes in subjects with metabolic abnormalities, including NAFLD and sarcopenia. Our results also provide a basis for stimulating further research into the role of crosstalk between liver and muscle in predicting the long-term prognoses of patients with NAFLD and/or sarcopenia.

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Author contributions

All authors did the conception and/or design; the acquisition, analysis, and/or interpretation of data; the drafting and/or revising of the work; and the final approval of the manuscript.

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The authors of this manuscript certify that they comply with the ethical guidelines for authorship and publishing in the *Journal of Cachexia, Sarcopenia and Muscle*.[58]

Online supplementary material

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Table S1. Baseline clinical characteristics according to nonalcoholic fatty liver disease and advanced fibrosis
Table S2. Baseline clinical characteristics according to survival status
Table S3. Baseline clinical characteristics according to nonalcoholic fatty liver disease and sarcopenia

Table S4. Hazard ratios for mortality according to nonalcoholic fatty liver disease and sarcopenia in the fully adjusted model (Model 4 in Table 3)

Table S5. Hazard ratios for cardiovascular disease-specific and cancer-specific mortality according to nonalcoholic fatty liver disease and sarcopenia in the fully adjusted model (Model 4 in Table 3)

Conflict of interest

The authors have no conflicts of interest to disclose.

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