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ORIGINAL ARTICLE—LIVER, PANCREAS, AND BILIARY TRACT

Increase in the skeletal muscle mass to body fat mass ratio predicts the decline in transaminase in patients with nonalcoholic fatty liver disease

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Abstract

Background The aim of this retrospective study was to determine the effect of skeletal muscle and body fat on liver function in patients with nonalcoholic fatty liver disease (NAFLD) diagnosed by liver biopsy.

Methods Among the 219 patients with NAFLD enrolled in this study was a cohort of 139 patients who had their body composition measured with Inbody720 at baseline and at $\geq$ 1 year postbaseline, to elucidate the relationship between liver function and changes in skeletal muscle and body fat mass. Multivariate analysis was used to identify factors influencing low skeletal muscle mass index (SMI, defined as 7 kg/m^2 in men, and 5.7 kg/m^2 in women) and the skeletal muscle mass to body fat mass ratio (SF ratio).

Results Of the 219 patients enrolled, 27 (12.3%) had a low SMI. Patient age (> 70 years) and female gender were identified as risk factors for low SMI. Hepatic fibrosis was not associated with SMI. In the cohort followed up at baseline and 12 months later, transaminase activity, body fat mass, and SMI significantly decreased over time. Changes in the SF ratio were significantly associated with changes in liver function. An increase in the SF ratio [hazard ratio (HR) 10.99 in men, 6.849 in women] was a predictor of reduced ALT, independent of age and other backgrounds.

Conclusions In the patients with NAFLD, SMI was decreased, even in the early stages of NAFLD. Therapeutic strategies for NAFLD require a reduction in body fat mass and the maintenance of skeletal muscle is also needed.

Keywords Body fat mass · NAFLD · SMI · SF ratio

Abbreviations

ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
BFMI	Body fat mass index
BMI	Body mass index
BMR	Basal metabolic rate
CI	Confidence interval
FPG	Fasting plasma glucose
GGT	γ Glutamyl transpeptidase
HbA1c	Hemoglobin A1c
HDL-C	High-density lipoprotein cholesterol
HR	Hazard ratio
SF ratio	SMI to BFMI ratio
LDL-C	Low-density lipoprotein cholesterol
NAFLD	Nonalcoholic fatty liver disease
NAS	NAFLD activity score
NASH	Nonalcoholic steatohepatitis
SMI	Skeletal mass index

Introduction

Nonalcoholic fatty liver disease (NAFLD) is currently the most common liver disease worldwide, affecting an estimated 25% of the adult population [1]. NAFLD is a disease covering a wide spectrum, ranging from nonalcoholic fatty

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liver (NAFL), which is usually a benign condition, to nonalcoholic steatohepatitis (NASH), which can sometimes lead to liver cirrhosis or hepatocellular carcinoma without significant alcohol consumption [2, 3]. The prevalence of NAFLD is increasing rapidly because of Western dietary patterns and a lack of exercise. NAFLD is also a well-known risk factor for type 2 diabetes mellitus, chronic kidney disease, and cardiovascular disease.

Although several factors, such as a habitually high-calorie diet, low levels of physical activity, elevated oxidative stress, and genetics can contribute to the pathogenesis of NAFLD, insulin resistance is one of the most pivotal mechanisms underpinning NAFLD progression. Insulin resistance is induced by liver dysfunction as well as skeletal muscle and body fat disorders which play a key role in glucose and lipid metabolism. Such perturbations are often closely related to the onset and progression of NAFLD. Sarcopenia, characterized by a decline in skeletal muscle and low muscle strength, affects clinical outcomes, including quality of life, infection rate, and survival in patients with cirrhosis [4–6]. In general, idiopathic sarcopenia is referred to as primary (or age-related) *sarcopenia,* whereas the etiologic basis of secondary *sarcopenia* relates to other diseases, such as chronic liver disease, renal disease, inflammatory disease, and malignant tumors. In European populations, approximately 0.5–1.0% of skeletal muscle mass is lost per year after the age of 30 years, with the rate of decline dramatically accelerating after age 65 [7]. In Japan and Western countries, secondary sarcopenia reportedly occurs in 40–70% of patients with cirrhosis [8, 9].

Abnormal body composition, which includes obesity-related body fat mass and reduced skeletal muscle mass, is estimated to be associated with the progression of NAFLD. Therapeutic strategies for NAFLD need to focus on better managing these components of body composition.

The aim of this study was to determine (1) the association between skeletal muscle mass and NAFLD, and (2) the effect of body composition on the liver function of patients with NAFLD in Japan.

Methods

Patients

From January 2014 to October 2017, 347 patients who were consecutively diagnosed with NAFLD on the basis of liver biopsy findings of steatosis in $\geq$ 5% of hepatocytes—in the absence of other liver diseases such as viral hepatitis, autoimmune hepatitis, and drug-induced liver disease—were screened for study inclusion. Patients who were consuming more than 20 g of alcohol per day or for whom there was evidence of decompensated liver cirrhosis or hepatocellular carcinoma were excluded from the study. Among the patients who underwent a liver biopsy, 128 were excluded because of a lack of body composition data. A total of 219 patients were enrolled in the study. A follow-up cohort comprised 139 patients who underwent serial body composition tests 12 months after baseline. All patients provided written informed consent at the time of liver biopsy, and the study was conducted in accordance with the Declaration of Helsinki. This study protocol was approved by the institution's human research committees.

Laboratory and clinical parameters

Venous blood samples were collected in the morning after a 12-h overnight fast. Laboratory assays included blood cell counts and measurements of serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), γ glutamyl transpeptidase (GGT), total cholesterol, triglycerides, fasting plasma glucose (FPG), and type IV collagen 7 s. Hemoglobin A1c (HbA1c) was assayed using high-performance liquid chromatography and expressed in National Glycohemoglobin Standardization Program (NGSP) units (%). Parameters were measured with standard clinical chemistry laboratory techniques. Body mass index (BMI) was calculated as weight (kg)/[height (m)]^2. Type 2 diabetes mellitus (T2DM) was diagnosed according to the Report of the Expert Committee on the Diagnosis and Classification of Diabetes Mellitus or confirmed on the basis of patients being prescribed antihyperglycemic T2DM agents. Patients with serum cholesterol concentrations $>$ 220 mg/dL, triglyceride concentrations $>$ 160 mg/dL, or who were prescribed antidyslipidemia agents were considered dyslipidemic.

Liver histology

All enrolled patients underwent an ultrasound-guided percutaneous liver biopsy. Paraffin-embedded liver sections were prepared for hematoxylin and eosin or Masson's trichrome staining. Specimens were evaluated by 2 hepatic pathologists (Y.S. and N.M.) who were blinded to the clinical findings. An adequate liver biopsy sample was defined as a specimen $>$ 1.5 cm in length and/or with more than 11 portal tracts. The criteria for a diagnosis of NASH was (1) any degree of steatosis in addition to centrilobular ballooning and/or Mallory-Denk bodies or (2) any degree of steatosis along with centrilobular pericellular/perisinusoidal fibrosis. Patients with liver biopsy specimens showing simple steatosis or steatosis with nonspecific inflammation were identified as the NAFL cohort. Specimens with steatosis thresholds of $<$ 5, 5–33, $>$ 33–66, and $>$ 66% were scored as grades 0, 1, 2, and 3,

respectively. Histological grade and stage were scored as described. Necroinflammatory grades of NASH were defined as grade 1 (mild), grade 2 (moderate), and grade 3 (severe) based on the extent of hepatocellular steatosis, ballooning, and inflammation (acinar and portal). The severity of hepatic fibrosis (stage) was scored as follows: stage 1, zone 3 perisinusoidal fibrosis; stage 2, zone 3 perisinusoidal fibrosis with portal fibrosis; stage 3, zone 3 perisinusoidal fibrosis and portal fibrosis with bridging fibrosis; and stage 4, cirrhosis [10–12].

Body composition

We analyzed the body composition of participants using the Inbody720 multifrequency impedance body composition analyzer (Inbody Japan, Tokyo, Japan). Body composition was measured in kilograms and basal metabolic rate (BMR) was calculated using the Cunningham formula of BMR = 21.6 × fat-free mass (kg) + 370. Skeletal muscle index (SMI) was calculated as skeletal muscle mass (kg)/[height (m)]^2. Body fat mass index (BFMI) was calculated as body fat mass (kg)/[height (m)]^2. As indicated in the sarcopenia diagnostic criteria of the Japan Society of Hepatology [13], we defined a low SMI as < 7.0 kg/m^2 in men and < 5.7 kg/m^2 in women.

Statistical analysis

Results are presented as numbers for qualitative data or as medians for quantitative data. The distribution of subject characteristics was assessed using the Chi-square test or Fisher's exact probability test, as appropriate. Logistic analysis was used to determine independent predictive factors associated with reduction in ALT in follow-up cohort. The hazard ratio (HR) and 95% confidence interval (CI) were also calculated. We performed multivariate logistic regression analysis with gender, age, complicating hypertension, hyperlipidemia, T2DM, serum level of AST, ALT, GGT, platelet count, hepatic fibrosis stage, and NAS to clarify the predictive factors associated with lowering SMI. The multivariate analysis to identify the predictive factors associated with reduction in ALT were performed with age, complicating hypertension, hyperlipidemia, T2DM, serum level of GGT, platelet count, hepatic fibrosis stage, NAS, and change in SF ratio. Statistical comparisons were performed using SPSS Ver.25 software (SPSS Inc., Chicago, IL). All P values < 0.05 were calculated using a two-tailed test and were considered significant.

Results

Patient characteristics

Baseline clinical characteristics of patients, as well as laboratory and histological data, are shown in Table 1. Total of 90 patients (41.1%) received the oral medication for T2DM, 98 patients (44.8%) received the oral medication for dyslipidemia, and 11 patients (5.0%) received the Vitamin E. The details of medication at baseline are described in Supplement Table 1. In this study of 219 patients, a cohort of 155 (70.8%) patients was diagnosed with NASH. T2DM was diagnosed in 61.9% of patients in the NASH group compared with 23.4% of patients in the NAFL group ($P < 0.001$). Of the 155 patients with NASH, 15 (9.7%) were stage 0, 59 (38.1%) were stage 1, 48 (31.0%) were stage 2, 19 (12.3%) were stage 3, and 14 (9.0%) had cirrhosis (stage 4). AST, ALT, GGT, FGP, HbA1c, and type IV collagen 7 s concentrations in serum, as well as BFMI, were significantly higher in patients with NASH than in patients with NAFL. In contrast, the platelet count was significantly lower in patients with NASH than in patients with NAFL. SMI was not significantly different between patients with NASH and NAFL, whereas SF ratio was significantly higher in patients with NAFL than in patients with NASH.

Prevalence of low SMI in patients with NAFLD

Patients were stratified on the basis of low and normal SMI (Table 2). SMI was low in 27 (12.3%) patients and normal in 192 (87.7%) patients. The low SMI group included more patients who were female, older, and had higher serum concentrations of AST and triglycerides than the normal SMI group. BMI, BFMI, SMI, total body water, and BMR were significantly lower in the group of patients with a lower SMI as opposed to a normal SMI.

The results for SMI, which was evaluated based on gender, age, and stage of hepatic fibrosis, are shown in Fig. 1. SMI decreased gradually with age in men. In women, SMI was significantly different between patients < 60 versus ≥ 60 years ($P = 0.028$). SMI was not correlated with stage of hepatic fibrosis. The prevalence of a low SMI for stages was as follows: stage 0, 14.8%; stage 1, 10.1%; stage 2, 6.1%; stage 3, 21.1%; and stage 4, 21.4%.

Factors associated with a low SMI

The multivariate analyses with gender, age, complicating hypertension, hyperlipidemia, T2DM, serum level of AST, ALT, GGT, platelet count, hepatic fibrosis stage, and NAS

Table 1 Demographic characteristics, as well as laboratory and histological data, for patients with nonalcoholic fatty liver disease[†]

Variables	Total $N = 219$	NAFL $n = 64$	NASH $n = 155$	P value[‡]
Female gender	119 (54.3%)	31 (48.4%)	88 (56.8%)	0.297
Age, years	58 (17–84)	55.5 (17–80)	61 (17–84)	0.089
Type 2 diabetes mellitus	111 (50.7%)	15 (23.4%)	96 (61.9%)	**<0.001**
Albumin, g/dL	4.4 (3.2–5.2)	4.4 (3.8–5.2)	4.4 (3.2–5.2)	0.702
AST, IU/L	42 (12–192)	33 (16–81)	46 (12–192)	**<0.001**
ALT, IU/L	52 (7–237)	44.5 (13–191)	56 (7–237)	**0.009**
GGT, IU/L	55 (14–716)	44.5 (15–716)	59 (14–374)	**0.015**
Platelet count, $\times 10^3/\mu L$	213 (76–444)	238 (92–444)	208 (76–412)	**0.027**
Total cholesterol, mg/dL	201 (123–335)	205 (133–288)	200 (123–355)	0.771
Triglycerides, mg/dL	139 (43–923)	142 (43–530)	139 (49–923)	0.638
LDL cholesterol, mg/dL	121.5 (45–259)	124 (66–185)	118 (45–259)	0.193
HDL cholesterol, mg/dL	52 (22–105)	53 (31–93)	52 (22–105)	0.770
FPG, mg/dL	108 (61–374)	101 (78–325)	109.5 (61–374)	**0.004**
HbA1c, %	6.3 (5.0–11.1)	5.9 (5.2–9.2)	6.5 (5.0–11.1)	**<0.001**
Type IV collagen 7 s, ng/mL	5.3 (2.5–12)	5.0 (2.5–10)	5.5 (2.8–12)	**0.005**
Liver function				
Steatosis, 0/1/2/3	0/64/117/38	0/26/26/1 2	0/38/91/26	**0.032**
Lobular inflammation, 0/1/2/3	9/117/78/15	9/49/5/1/63	0/68/73/14	**<0.001**
Hepatocyte ballooning, 0/1/2	63/84/72	63/1/0	0/83/72	**<0.001**
Fibrosis stage, 0/1/2/3/4	68/69/49/19/14	53/10/1/0/0	15/59/48/19/14	**<0.001**
Body composition				
BMI, kg/m^2	27.2 (19.2–44.2)	26.2 (19.4–44.2)	27.4 (19.2–43.9)	0.054
BFMI, kg/m^2	9.4 (1.9–23.2)	8.1 (1.9–23.2)	10.0 (4.2–20.1)	**0.010**
SMI, kg/m^2	7.29 (4.86–10.43)	7.29 (4.89-10.07)	7.29 (4.86–10.43)	0.689
Total body water, L	33.2 (20.9–54.5)	33.2 (20.9–50.2)	33.6 (21.4–54.5)	0.688
Basal metabolic rate, kcal	1339 (986–1972)	1339 (986–1851)	1339 (998–1972)	0.653
SF ratio	0.77 (0.25–3.76)	0.88 (0.25–3.76)	0.72 (0.38–1.70)	**0.015**

ALT alanine aminotransferase, *AST* aspartate aminotransferase, *BFMI* body fat mass index, *BMI* body mass index, *FPG* fasting plasma glucose, *GGT* γ glutamyl transpeptidase, *HbA1c* hemoglobin A1c, *HDL-C* high-density lipoprotein cholesterol, *LDL-C* low-density lipoprotein cholesterol, *NAFL* nonalcoholic fatty liver, *NASH* nonalcoholic steatohepatitis, *SMI* skeletal muscle mass index, *SF ratio* SMI to BFMI ratio

[†]Results are presented as a number (n) or n (%) for qualitative data or as median (range) for quantitative data

[‡]Bold font for P values indicates less than 0.05

were performed to identify the risk factors associated with a low SMI in patients with NAFLD and revealed that the adjusted hazard ratio (HR) for women was 2.949 (95% CI 1.101–9.979; $P = 0.031$) and for the elderly ($\geq$ 70 years) the HR was 5.114 (95% CI 1.259–20.78, $P = 0.023$; Table 3). The prevalence of a low SMI was 16.8% for women, 29.5% for men and women $\geq$ 70 years, and 35.7% in women $\geq$ 70 years in the NAFLD cohort.

Changes in liver function and body composition in the follow-up cohort

A total of 139 patients underwent body composition tests at baseline and 12 months postbaseline. We performed only nutrition and exercise therapy, and no patients were added oral medication in follow-up period. The serum concentrations of AST, ALT, GGT, and HbA1c significantly decreased by the 12-month follow-up. Not only BFMI but also SMI significantly decreased by the 12-month follow-up (Table 4). Changes in the liver function parameters AST and ALT were negatively correlated with the ratio of SMI:BFMI (SF ratio) (Supplement Figure 1). Patients with an increased SF ratio exhibited a larger reduction in ALT ($-$ 21.5 IU/L) than patients without an increased SF ratio ($-$ 1.0 IU/L, $P < 0.001$). Even in the BMI increased group, the patients with increased SF ratio tended to show greater reduction in ALT ($-$ 20.0 IU/L), than the patients with decreased SF ratio (0.5 IU/L, $P = 0.053$) (Fig. 2).

To estimate the risk factors associated with reduction in ALT, we examined the correlation of patients'

Table 2 Patient characteristics according to skeletal muscle index[†]

Variables	SMI		*P* value[‡]
	Low *n* = 27	Normal *n* = 193	
Gender, female	20 (74.1%)	99 (51.6%)	**0.038**
Age, years	68 (21–84)	58 (17–80)	**0.015**
Type 2 diabetes mellitus	9 (33.3%)	55 (28.6%)	0.541
Albumin, g/dL	4.4 (3.6–5.2)	4.4 (3.2–5.2)	0.671
AST, IU/L	56 (24–135)	41 (12–192)	**0.018**
ALT, IU/L	73 (19–163)	51 (7–237)	0.067
GGT, IU/L	93 (18–716)	53 (14–374)	0.084
Platelet count, $\times 10^3/\mu L$	207 (95–377)	195 (76–444)	0.349
Total cholesterol, mg/dL	191 (144–278)	215 (123–355)	0.144
Triglycerides, mg/dL	157 (91–565)	138 (43–923)	**0.021**
LDL cholesterol, mg/dL	120 (66–169)	121.5 (45–259)	0.937
HDL cholesterol, mg/dL	52 (36–83)	52 (22–105)	0.693
FPG, mg/dL	107 (84–189)	108 (61–374)	0.546
HbA1c, %	6.2 (5.2–11.0)	6.3 (5.0–11.1)	0.801
Type IV collagen 7 s, ng/mL	5.5 (3.4–12.0)	5.3 (2.5–12.0)	0.112
Liver function			
Steatosis, 0/1/2/3	0/11/11/5	0/53/106/33	0.309
Lobular inflammation, 0/1/2/3	2/15/6/4	7/102/72/11	0.161
Hepatocyte ballooning, 0/1/2	8/7/12	55/77/60	0.286
Fibrosis stage, 0/1/2/3/4	10/7/3/4/3	58/62/46/15/11	0.307
Body composition			
BMI, kg/m^2	21.9 (19.2–30.8)	27.7 (19.4–44.2)	**<0.001**
BFMI, kg/m^2	7.6 (4.2–21.8)	9.9 (1.9–23.2)	**<0.001**
SMI, kg/m^2	13.2 (4.9–6.9)	20.0 (5.8–10.4)	**<0.001**
Total body water, L	25.5 (20.9–34.1)	35.0 (23.0–54.5)	**<0.001**
Basal metabolic rate, kcal	1117 (986–1372)	1394 (1044–1972)	**<0.001**

ALT alanine aminotransferase, *AST* aspartate aminotransferase, *BFMI* body fat mass index, *BMI* body mass index, *FPG* fasting plasma glucose, *GGT* γ glutamyl transpeptidase, *HbA1c* hemoglobin A1c, *HDL-C* high-density lipoprotein cholesterol, *LDL-C* low-density lipoprotein cholesterol, *SMI* skeletal muscle mass index

[†]Results are presented as a number (n) or mean *n* (%) for qualitative data or as median (range) for quantitative data

[‡]Bold font for *P* values indicates less than 0.05

backgrounds with reduction in ALT. We performed multivariate logistic regression analysis with age, complicating hypertension, hyperlipidemia, T2DM, serum level of GGT, platelet count, hepatic fibrosis stage, NAFLD activity score (NAS), and change in SF ratio in each sex. The multivariate analysis revealed that an increased SF ratio (HR of 10.99, $P = 0.021$ in men, HR of 6.849, $P = 0.015$ in women) was an independent predictive factor for a reduction in ALT in each sex, and the pathological findings having no discernable effect (Table 5). To compare the impact to liver function, we also performed multivariate logistic regression analysis with change in BMI instead of SF ratio (Supplement Table 2). The HR of increased SF ratio was greater than decreased BMI (HR 4.215, $P = 0.031$) in women. The change in BMI was not detected as a predictive factor in men.

Discussion

We investigated the SMI of Japanese patients with NAFLD by monitoring body composition. The prevalence of a low SMI was 12.3% in this study. A previous study investigating the prevalence of sarcopenia showed that 17.9% of patients with NAFL and 35.0% of patients with NASH had sarcopenia [14]. Another study by Lee et al. reported that 12.2% of patients with NAFLD were diagnosed with sarcopenia [15]. These two studies determined that the severity of hepatic fibrosis was associated with sarcopenia.

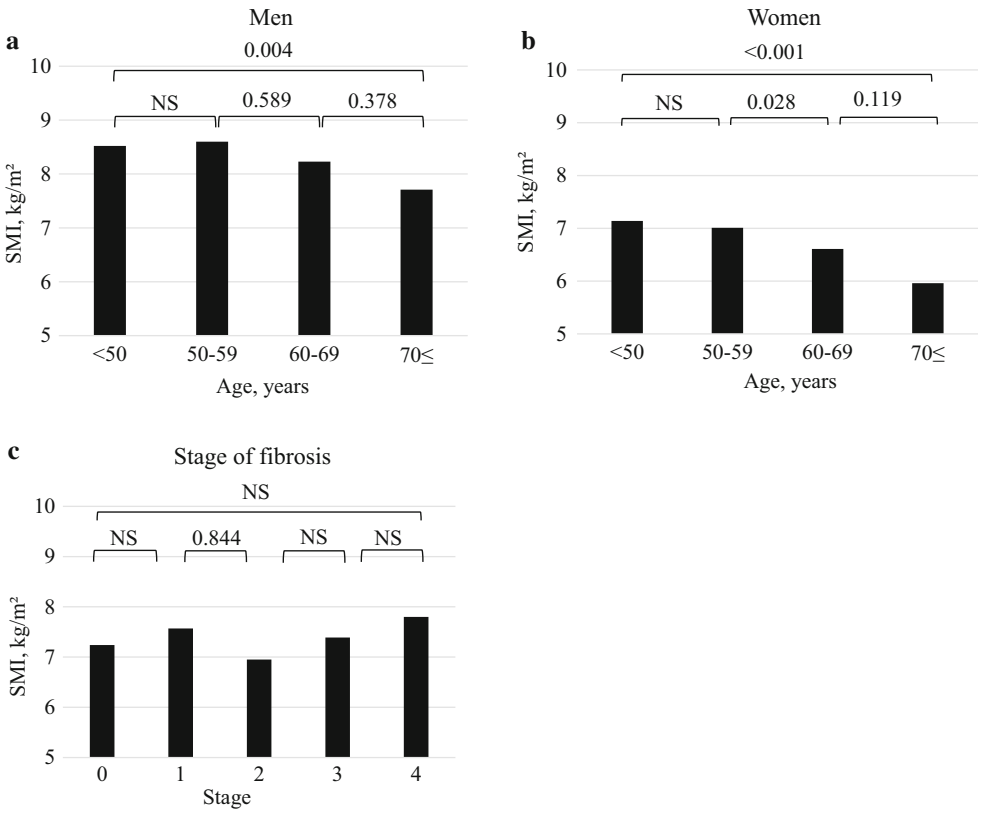

Fig. 1 Skeletal muscle mass index (SMI) evaluated according to gender, age, and stage of hepatic fibrosis

In the present study, there was no correlation between SMI and histological features. Even among patients who did not have hepatic fibrosis, 14.5% of them met the low SMI criteria. The reason for this discrepancy is unclear; however, the definitions for sarcopenia, NAFLD, and the size and ethnicity of the population may be contributing that. The existence of patients with a low SMI during an early stage of NAFLD suggests that monitoring of SMI may be prudent at an early stage especially they are elderly patients.

The etiology for the reduction in SMI was different between men and women. The SMI decrease occurred gradually with progressive aging in men, whereas there was a significant decrease after the age of 60 years in women. One plausible hypothesis for these disparate results is that menopause affects the body composition of women. Menopause precipitates a decline in estrogen concentrations, and an increase in visceral adiposity, as well as a decrease in bone density, muscle mass, and muscle strength [16, 17]. The effect of menopause on skeletal muscle mass can occur directly as a consequence of estrogen receptors on muscle cells, and indirectly via an increase in circulating inflammatory cytokines [18]. Several studies also report a vitamin D deficiency can be associated with sarcopenia, especially in elderly women [19–21].

In this retrospective study, we identified the importance of the SF ratio in the pathology of NAFLD. After 12 months of follow-up, serum concentrations of AST, ALT, and BFMI decreased significantly. It is also worth mentioning that SMI declined significantly by 12 months. The changes in liver function were significantly associated with changes in the SF ratio. Targeting visceral fat stores using medication and/or exercise therapy to reduce them is considered essential to therapeutic approaches aiming to manage NAFLD [22–24]. A reduction in visceral fat accumulation is associated with improvements in insulin resistance, hypertension, dyslipidemia, and systemic chronic low-grade inflammation, and can decrease the risk of atherosclerosis and cardiovascular disease [25–27]. Because East Asian populations, including the Japanese, generally have a higher percentage of body fat than Caucasians of the same age, gender, and BMI [28–31], body fat or BMI is often the target of therapy.

Loss of skeletal muscle mass, especially when it is associated with sarcopenia, can be an independent risk factor for infection, a lower quality of life, and may even be prognostic in patients with cirrhosis [4, 5, 32, 33]. The effect of a low SMI on the pathology of NAFLD in the absence of cirrhosis is still a matter of debate. Interestingly, *sarcopenic obesity*—defined as a high fat mass and a low muscle mass—has been receiving attention as a risk factor for the impairment of physical activity and worse clinical

Table 3 Factors associated with lowering SMI in patients with NAFLD as identified by multivariate analyses

Variables	Category	Multivariate analysis	
		HR (95% CI)[†]	*P* value[‡]
Gender	1: male	2.949 (1.101–9.979)	**0.031**
	2: female		
Age, years	1: < 70	5.114 (1.259–20.78)	**0.023**
	2: ≥ 70		
Hypertension	1: no	0.333 (0.097–1.146)	0.081
	2: yes		
Hyperlipidemia	1: no	0.843 (0.256–2.781)	0.779
	2: yes		
Diabetes Mellitus	1: no	0.847 (0.260–2.757)	0.782
	2: yes		
AST, IU/L	1: < 40	1.370 (0.254–7.394)	0.714
	2: ≥ 40		
ALT, IU/L	1: < 40	2.927 (0.494–17.32)	0.237
	2: ≥ 40		
GGT, IU/L	1: < 70	1.932 (0.605–6.168)	0.266
	2: ≥ 70		
Platelet count, × 10^3/μL	1: > 200	1.040 (0.280–3.855)	0.954
	2: ≤ 200		
Fibrosis stage	1: 0, 1, 2	3.358 (0.623–18.09)	0.159
	2: 3, 4		
NAS	1: < 5	0.626 (0.144–2.728)	0.533
	2: ≥ 5		

[†]Estimated using Cox proportional hazards regression analysis

[‡]Bold font for *P* values indicates less than 0.05

ALT alanine aminotransferase, *AST* aspartate aminotransferase, *CI* confidence interval, *GGT* γ glutamyl transpeptidase, *HR* hazard ratio, *NAS* NAFLD activity score

outcomes compared with sarcopenia or obesity alone [34, 35]. Shida et al. suggest that a reduction in skeletal muscle mass may synergistically increase visceral fat in overweight subjects with NAFLD [36]. Leptin and inflammatory cytokines such as IL-6 are thought to play a major role in this synergy. Leptin stimulates fat degradation in skeletal muscle and improves insulin sensitivity [37]. In patients with sarcopenic obesity, serum leptin levels are reportedly higher than in patients with sarcopenia or visceral fat accumulation only [38]. Leptin resistance leads to a reduction in muscle mass and stimulates fat accumulation in muscles via an AMPK pathway-mediated reduction in muscle fatty acid oxidation. Serum IL-6 was reported to be high in subjects with sarcopenic obesity [39, 40]. The report by Shida et al. indicates that changes in body composition were associated with a change in adipokines, myokines, and hepatokines [36], thus supporting our result that changes in the SF ratio were associated with reduced ALT. The reduction in ALT concentrations in patients with an increased SF ratio after 12 months was significantly greater (− 21 IU/L) than for those patients with a decreased SF ratio (− 1.5 IU/L; $P < 0.001$). In the BMI decreasing group, the diminution of ALT was significantly greater in patients with increased SF ratio than in patients with decreased SF ratio (p = 0.005, Fig. 2). On the other hand, even the BMI increased, the patients with increased SF ratio showed reduction in ALT (− 20.0 IU/L) as much as the patients with decreased BMI and increased SF ratio (Fig. 2). From the standpoint of exercise therapy, there was no significant difference between aerobic and resistance exercise on changes in BMI, serum ALT, and hepatic steatosis [41]. Resistance exercise was reported to improve NAFLD in conjunction with consuming less energy, and patients unable to tolerate or adequately participate in aerobics exercise may benefit from performing resistance exercise as part of their therapeutic regimen [41].

This study has several limitations. It is a retrospective study conducted at a single center and the number of patients studied was not large. A multicenter, prospective study enrolling a larger number of patients will be required to draw firm conclusions. Because of the low prevalence of

Table 4 Demographic characteristics as well as laboratory and histological data for patients in the followed up cohort[†]

Variables	Patients with NAFLD		*P* value[‡]
	Baseline *n* = 139	12 months *n* = 139	
Age, years	58 (17–80)	–	–
Albumin, g/dL	4.4 (3.5–5.2)	4.4 (3.0–7.7)	0.726
AST, IU/L	42 (12–192)	32 (15–139)	**<0.001**
ALT, IU/L	54 (37–202)	35 (13–262)	**<0.001**
GGT, IU/L	58 (14–355)	45 (14–332)	**<0.001**
Platelet count, $\times 10^3/\mu L$	216 (76–444)	220 (51–419)	0.704
Total cholesterol, mg/dL	205 (123–355)	197.5 (55–280)	0.769
Triglycerides, mg/dL	131 (49–565)	134 (40–781)	0.459
LDL cholesterol, mg/dL	126 (45–259)	122 (47–217)	0.704
HDL cholesterol, mg/dL	53 (22–105)	52 (25–94)	0.258
FPG, mg/dL	106 (61–374)	110 (84–287)	0.357
HbA1c, %	6.4 (5.0–11.1)	6.1 (4.9–10.5)	**0.006**
Body composition			
BMI, kg/m^2	27.2 (19.5–40.4)	26.56 (19.2–40.6)	**<0.001**
BFMI, kg/m^2	24.9 (19.5–40.4)	23.3 (3.0–20.6)	**<0.001**
SMI, kg/m^2	7.28 (4.89–9.87)	7.20 (4.99–9.75)	**0.001**
Total body water, L	33.2 (20.9–54.5)	32.1 (21.6–53.7)	**<0.001**
Basal metabolic rate, kcal	1339 (986–1972)	1311 (1007–1944)	**<0.001**
SF ratio	0.74 (0.38-2.08)	0.77 (0.40-2.41)	**0.018**

AST aspartate aminotransferase, *ALT* alanine aminotransferase, *FPG* fasting plasma glucose, *GGT* γ glutamyl transpeptidase, *HDL-C* high-density lipoprotein cholesterol, *LDL-C* low-density lipoprotein cholesterol, *NAFLD* nonalcoholic fatty liver disease

[†]Results are the median (range) for quantitative data

[‡]Bold font for *P* values indicates less than 0.05

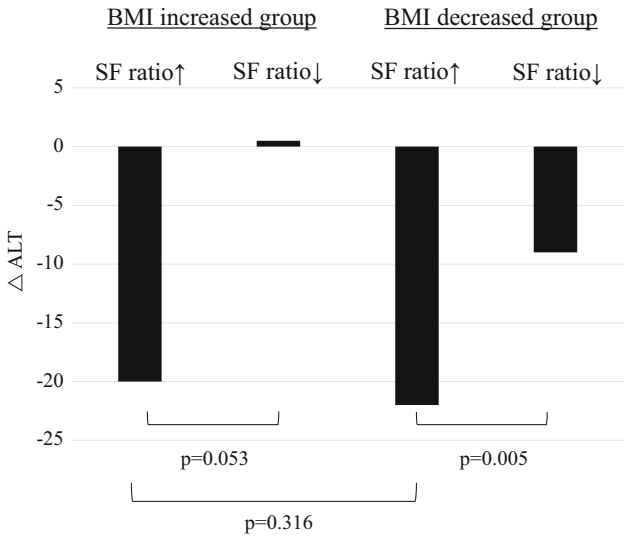

Fig. 2 The change in ALT according to change in BMI and SF ratio. *Delta* change from baseline to 12 months

advanced fibrosis (> stage 3, 15.1%), there is a possibility that the lowering SMI that was seen in the elderly cases in this study is simply exhibits general phenomenon in aging. Next, we did not measure muscle strength in this study. Because sarcopenia is diagnosed according to the loss of skeletal muscle mass and muscle strength, it is possible to overlook the size and number of muscle fiber decreases. A major strength of this study is that all enrolled patients were diagnosed by liver biopsy. A second strength relates to the use of bioelectrical impedance analysis to evaluate body composition. The previous report that compared the bioelectrical impedance analysis with dual X-ray absorptiometry showed that the ability of estimate the amount of skeletal muscle mass was equal in both methods [42]. Furthermore, the bioelectrical impedance analysis is a simple and noninvasive method rather than dual X-ray absorptiometry method including computed tomography or magnetic resonance imaging. From the economical point of view, the bioelectrical impedance analysis has also priority to Dual X-ray absorptiometry method.

In conclusion, patients with NAFLD were potentially susceptible to a low SMI, even during the early stage of disease development, and the changes in the SF ratio were correlated with parameters for liver function. Our results suggest that it would be prudent to evaluate the body composition of patients with NAFLD and incorporate the

Table 5 Factors associated with amelioration of ALT in patients with NAFLD

Variables[a]	Category	Multivariate analysis	
		HR (95% CI)[†]	*P* value[‡]
a. Men *n* = 57			
Age, years	1: < 70	0.384 (0.016–9.175)	0.554
	2: ≥ 70		
Hypertension	1: no	3.179 (0.309–32.73)	0.331
	2: yes		
Hyperlipidemia	1: no	1.181 (0.220–6.337)	0.846
	2: yes		
Diabetes Mellitus	1: no	0.358 (0.051–2.527)	0.303
	2: yes		
GGT, IU/L	1: < 70	5.486 (1.034–29.10)	**0.046**
	2: ≥ 70		
Platelet count, $x10^3/\mu L$	1: > 200	0.766 (0.109–5.395)	0.789
	2: ≤ 200		
Fibrosis stage	1: 0, 1, 2	0.653 (0.036–12.00)	0.774
	2: 3, 4		
NAS	1: < 5	2.796 (0.325–24.07)	0.349
	2: ≥ 5		
Change in SF ratio	1: decreased	10.99 (1.437–83.33)	**0.021**
	2: increased		
b. Women n = 82			
Age, years	1: < 70	0.635 (0.106–3.798)	0.619
	2: ≥ 70		
Hypertension	1: no	0.629 (0.152–2.607)	0.523
	2: yes		
Hyperlipidemia	1: no	4.407 (0.996–19.51)	0.051
	2: yes		
Diabetes Mellitus	1: no	1.017 (0.284–3.648)	0.979
	2: yes		
GGT, IU/L	1: < 70	0.659 (0.172–2.524)	0.543
	2: ≥ 70		
Platelet count, $\times 10^3/\mu L$	1: > 200	5.194 (1.015–26.59)	**0.048**
	2: ≤ 200		
Fibrosis stage	1: 0, 1, 2	0.311 (0.050–1.951)	0.212
	2: 3, 4		
NAS	1: < 5	0.568 (0.104–3.109)	0.514
	2: ≥ 5		
Change in SF ratio	1: decreased	6.849 (1.443–32.26)	**0.015**
	2: increased		

GGT γ glutamyl transpeptidase, *NAS* NAFLD activity score, *CI* confidence interval, *HR* hazard ratio, *NAFLD* nonalcoholic fatty liver disease, *SF* ratio of skeletal mass index to body fat mass index

[†]Estimated using Cox proportional hazards regression analysis

[‡]Bold font for *P* values indicates less than 0.05

monitoring of these indices to reduce the risk of sarcopenia and a worse prognosis.

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Compliance with ethical standards

Conflict of interest The authors do not have any disclosures to report.

References

1. Younossi ZM, Koenig AB, Abdelatif D, et al. Global epidemiology of nonalcoholic fatty liver disease-meta-analytic assessment of prevalence, incidence, and outcomes. Hepatology. 2016;64:73–84.
2. Chalasani N, Younossi Z, Lavine JE, et al. The diagnosis and management of non-alcoholic fatty liver disease: practice guideline by the American Association for the Study of Liver Diseases, American College of Gastroenterology, and the American Gastroenterological Association. Hepatology. 2012;55:2005–23.
3. Watanabe S, Hashimoto E, Ikejima K, et al. Evidence based clinical practice guidelines for nonalcoholic fatty liver disease/nonalcoholic steatohepatitis. J Gastroenterol. 2015;50:364–77.
4. Montano-Loza AJ, Meza-Junco J, Prado CM, et al. Muscle wasting is associated with mortality in patients with cirrhosis. Clin Gastroenterol Hepatol. 2012;10:166–73 (**173.e1**).
5. Shiraki M, Nishiguchi S, Saito M, et al. Nutritional status and quality of life in current patients with liver cirrhosis as assessed in 2007–2011. Hepatol Res. 2013;43:106–12.
6. Huisman EJ, Trip EJ, Siersema PD, van Hoek B, van Erpecum KJ. Protein energy steatosis grades of 0, 1, 2, and 3, malnutrition predicts complications in liver cirrhosis. Eur J Gastroenterol Hepatol. 2011;23:982–9.
7. Kyle UG, Genton L, Hans D, Karsegard L, Slosman DO, Pichard C. Age-related differences in fat-free mass, skeletal muscle, body cell mass and fat mass between 18 and 94 years. Eur J Clin Nutr. 2001;55:663–72.
8. Scaglione S, Kliethermes S, Cao G, et al. The epidemiology of cirrhosis in the United States: a population-based study. J Clin Gastroenterol. 2015;49:690–6.
9. Hanai T, Shiraki M, Ohnishi S, et al. Rapid skeletal muscle wasting predicts worse survival in patients with liver cirrhosis. Hepatol Res. 2016;46:743–51.
10. Matteoni CA, Younossi ZM, Gramlich T, et al. Nonalcoholic fatty liver diseases: a spectrum of clinical and pathological severity. Gastroenterology. 1999;116:1413–9.
11. Kleiner DE, Brunt EM, Van Natta M, et al. Design and validation of a histological scoring system for nonalcoholic fatty liver disease. Hepatology. 2005;41:1313–21.
12. Brunt EM, Janney CG, Di Bisceglie AM, et al. Nonalcoholic steatohepatitis: a proposal for grading and staging the histological lesions. Am J Gastroenterol. 1999;94:2467–74.
13. Nishikawa H, Shiraki M, Hiramatsu A, et al. Japan Society of Hepatology guidelines for sarcopenia in liver disease (1st edition): recommendation from the working group for creation of sarcopenia assessment criteria. Hepatol Res. 2016;46:951–63.
14. Koo BK, Kim D, Joo SK, et al. Sarcopenia is an independent risk factor for non-alcoholic steatohepatitis and significant fibrosis. J Hepatol. 2017;66:123–31.
15. Lee YH, Kim SU, Song K, et al. Sarcopenia is associated with significant liver fibrosis independently of obesity and insulin resistance in nonalcoholic fatty liver disease: Nationwide surveys (KNHANES 2008-2011). Hepatology. 2016;63:776–86.
16. Messier V, Rabasa-Lhoret R, Barbat-Artigas S, et al. Menopause and sarcopenia: a potential role for sex hormones. Maturitas. 2011;68:331–6.
17. Anagnostis P, Dimopoulou C, Karras S, et al. Sarcopenia in postmenopausal women: is there any role for vitamin D? Maturitas. 2015;82:56–64.
18. Brown M. Skeletal muscle and bone: effect of sex steroids and aging. Adv Physiol Educ. 2008;32:120–6.
19. Visser M, Deeg DJ, Lips P. Low vitamin D and high parathyroid hormone levels as determinants of loss of muscle strength and muscle mass (sarcopenia): the Longitudinal Aging Study Amsterdam. J Clin Endocrinol Metab. 2003;88:5766–72.
20. Flicker L, Mead K, MacInnis RJ, et al. Serum vitamin D and falls in older women in residential care in Australia. J Am Geriatr Soc. 2003;51:1533–8.
21. Park S, Ham JO, Lee BK. A positive association of vitamin D deficiency and sarcopenia in 50 year old women, but not men. Clin Nutr. 2014;33:900–5.
22. Finelli C, Tarantino G. Is visceral fat reduction necessary to favour metabolic changes in the liver? J Gastrointestin Liver Dis. 2012;21:205–8.
23. Bouchi R, Nakano Y, Fukuda T, et al. Reduction of visceral fat by liraglutide is associated with ameliorations of hepatic steatosis, albuminuria, and micro-inflammation in type 2 diabetic patients with insulin treatment: a randomized control trial. Endocr J. 2017;64:269–81.
24. Houghton D, Thoma C, Hallsworth K, et al. Exercise reduces liver lipids and visceral adiposity in patients with nonalcoholic steatohepatitis in a randomized controlled Trial. Clin Gastroenterol Hepatol. 2017;15:96–102.
25. Yusuf S, Hawken S, Ounpuu S, et al. Obesity and the risk of myocardial infarction in 27,000 participants from 52 countries: a case–control study. Lancet. 2005;366:1640–9.
26. Fujimoto WY, Bergstrom RW, Boyko EJ, et al. Visceral adiposity and incident coronary heart disease in Japanese-American men. The 10-year follow-up results of the Seattle Japanese-American Community Diabetes Study. Diabetes Care. 1999;22:1808–12.
27. Britton KA, Massaro JM, Murabito JM, et al. Body fat distribution, incident cardiovascular disease, cancer, and all-cause mortality. J Am Coll Cardiol. 2013;62:921–5.
28. Hall AM, Kou K, Chen Z, et al. Evidence for regulated monoacylglycerol acyltransferase expression and activity in human liver. J Lipid Res. 2012;53:990–9.
29. Farrell GC, Chitturi S, Lau GK, Sollano JD, Asia-Pacific Working Party on NAFLD. Guidelines for the assessment and management of non-alcoholic fatty liver disease in the Asia-Pacific region: executive summary. J Gastroenterol Hepatol. 2007;22:775–7.
30. WHO Expert Consultation. Appropriate body-mass index for Asian populations and its implications for policy and intervention strategies. Lancet. 2004;363:157–63.
31. Alberti KG, Zimmet P, Shaw J, IDF Epidemiology Task Force Consensus Group. The metabolic syndrome—a new worldwide definition. Lancet. 2005;366:1059–62.
32. Merli M, Lucidi C, Giannelli V, et al. Cirrhotic patients are at risk for health care-associated bacterial infections. Clin Gastroenterol Hepatol. 2010;8:979–85.
33. Fujiwara N, Nakagawa H, Kudo Y, et al. Sarcopenia, intramuscular fat deposition, and visceral adiposity independently predict the outcomes of hepatocellular carcinoma. J Hepatol. 2015;63:131–40.
34. Kob R, Bollheimer LC, Bertsch T, et al. Sarcopenic obesity: molecular clues to a better understanding of its pathogenesis? Biogerontology. 2015;16:15–29.
35. Prado CM, Wells JC, Smith SR, et al. Sarcopenic obesity: a critical appraisal of the current evidence. Clin Nutr. 2012;31:583–601.
36. Shida T, Akiyama K, Oh S, et al. Skeletal muscle mass to visceral fat area ratio is an important determinant affecting hepatic conditions of non-alcoholic fatty liver disease. J Gastroenterol. 2017. https://doi.org/10.1007/s00535-017-1377-3.
37. Dyck DJ. Adipokines as regulators of muscle metabolism and insulin sensitivity. Appl Physiol Nutr Metab. 2009;34:396–402.

38. Kohara K, OchiM Tabara Y, et al. Leptin in sarcopenic visceral obesity: possible link between adipocytes and myocytes. PLoS ONE. 2011;6:e24633.
39. Cesari M, Kritchevsky SB, Baumgartner RN, et al. Sarcopenia, obesity, and inflammation—results from the trial of angiotensin converting enzyme inhibition and novel cardiovascular risk factors study. Am J Clin Nutr. 2005;82:428–34.
40. Schrager ME, Metter EJ, Simonsick E, et al. Sarcopenic obesity and inflammation in the InCHIANTI study. J Appl Physiol. 2007;102:919–25.
41. Hashida R, Kawaguchi T, Bekki M, et al. Aerobic vs. resistance exercise in non-alcoholic fatty liver disease: a systematic review. J Hepatol. 2017;66:142–52.
42. Yu SC, Powell A, Khow KS, Visvanathan R. The performance of five bioelectrical impedance analysis prediction equations against dual X-ray absorptiometry in estimating appendicular skeletal muscle mass in an adult Australian population. Nutrients. 2016;29(8):189.