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Moderate to Vigorous Physical Activity Volume Is an Important Factor for Managing Nonalcoholic Fatty Liver Disease: A Retrospective Study

Sechang Oh,[1] Takashi Shida,[2] Kazumasa Yamagishi,[3] Kiyoji Tanaka,[4] Rina So,[4] Takehiko Tsujimoto,[4] and Junichi Shoda[1]

Recently, the beneficial effects of increased physical activity (PA) on nonalcoholic fatty liver disease (NAFLD) in obese subjects were reported. However, the optimal strength and volume of PA in lifestyle modification to improve NAFLD pathophysiology and be recommended as an appropriate management of this condition are unclear. The primary goal of this retrospective study was to estimate the beneficial effects of a varying volume of moderate to vigorous intensity PA (MVPA) on the improvement of NAFLD. A total of 169 obese, middle-aged men were enrolled in a 12-week weight reduction program through lifestyle modification consisting of dietary restrictions plus aerobic exercise. Among these obese subjects, 40 performed MVPA for <150 min·wk^{-1}, 42 performed MVPA for 150-250 min·wk^{-1}, and 87 performed MVPA for >250 min·wk^{-1}. The subjects in the MVPA ≥250 min·wk^{-1} group, in comparison with those in the MVPA <250 min·wk^{-1} group, showed significantly attenuated levels of hepatic steatosis (−31.8% versus −23.2%). This attenuation was likely independent of the detectable weight reduction. MVPA for ≥250 min·wk^{-1} in comparison with that for <150 min·wk^{-1} led to a significant decrease in the abdominal visceral adipose tissue severity (−40.6% versus −12.9%), levels of ferritin (−13.6% versus +1.5%), and lipid peroxidation (−15.1% versus −2.8%), and a significant increase in the adiponectin levels (+17.1% versus +5.6%). In association with these changes, the gene expression levels of sterol regulatory element-binding protein-1c and carnitine palmitoyltransferase-1 in peripheral blood mononuclear cells also significantly decreased and increased, respectively. *Conclusion:* MVPA for ≥250 min·wk^{-1} as part of lifestyle management improves NAFLD pathophysiology in obese men. The benefits seem to be acquired through reducing inflammation and oxidative stress levels and altering fatty acid metabolism. (HEPATOLOGY 2015;61:1205-1215)

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With the increasing prevalence of overweight and obese middle-aged men in Japan, nonalcoholic fatty liver disease (NAFLD) is commonly diagnosed in daily clinical practice; analysis of data from

Abbreviations: ACC, acetyl-CoA carboxylase; ACS, acyl-CoA synthetase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; CAP, controlled attenuation parameter; CPT1, carnitine palmitoyltransferase I; FAS, fatty acid synthase; FFAs, free fatty acids; FPG, fasting plasma glucose; Fpn1, ferroportin-1; γGT, gamma glutamyl transpeptidase; HDL-C, high-density lipoprotein-cholesterol; HO1, heme oxygenase 1; HOMA-IR, insulin resistance by homeostasis model assessment; hs-CRP, high-sensitivity C-reactive protein; IL-6, interleukin 6; Mac2bp, mac-2 binding protein; MVPA, moderate to vigorous intensity physical activity; NAFLD, nonalcoholic fatty liver disease; NASH, nonalcoholic steatohepatitis; NQO1, NADH quinone oxidoreductase; Nrf2, nuclear factor E2-related factor 2; SAT, subcutaneous adipose tissue; SCD1, stearoyl-CoA desaturase-1; SREBP1c, sterol regulatory element-binding protein 1c; PBMCs, peripheral blood mononuclear cells; TBARS, thiobarbituric acid reactive substances; TEI, total daily energy intake; TF, total fat; TG, triglycerides; TNF-α, tumor necrosis factor alpha; VAT, visceral adipose tissue; WC, waist circumference.

From the [1]Division of Clinical Medicine, Faculty of Medicine, University of Tsukuba, Tsukuba, Ibaraki, Japan; [2]Doctoral Programs in Medical Sciences, Graduate School of Comprehensive Human Sciences, University of Tsukuba, Tsukuba, Ibaraki, Japan; [3]Division of Public Health Medicine, Faculty of Medicine, University of Tsukuba, Tsukuba, Ibaraki, Japan; [4]Division of Sports Medicine, Faculty of Health and Sports Science, University of Tsukuba, Tsukuba, Ibaraki, Japan;

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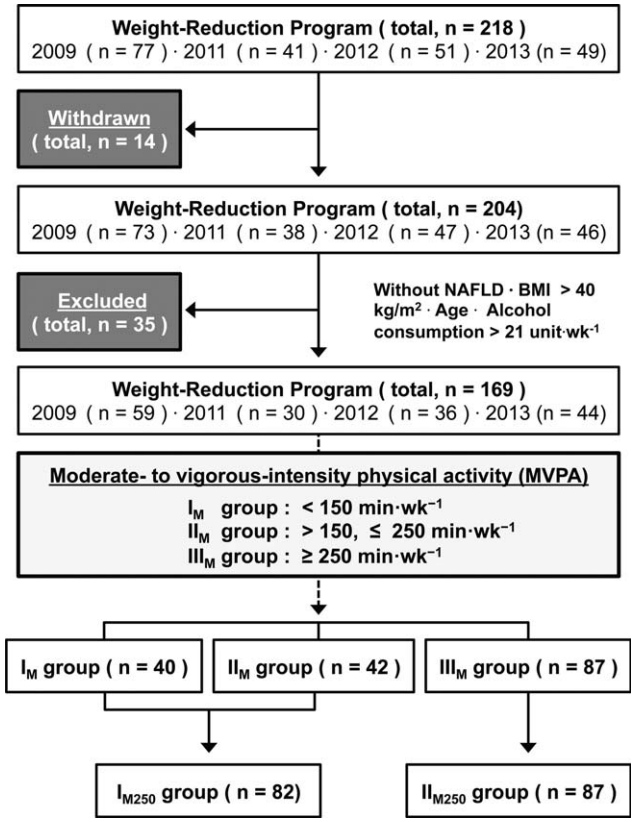

Fig. 1. Flow diagram of the enrollment and classification of study participants.

cross-sectional health examination surveys in the general population shows the prevalence of NAFLD in middle-aged men to be >40% in Japan.[1]

Weight reduction has been the only strategy established thus far to reduce hepatic lipid levels[2]; thus, dietary restrictions focused on weight reduction is recommended as the cornerstone for managing NAFLD.[3] With respect to physical activity (PA), recent reports on clinical outcomes have indicated that physical inactivity and low aerobic fitness are underlying reasons for the increased number of NAFLD cases,[4,5] and that both aerobic and resistance exercises have specific effects on NAFLD treatment in the absence of weight reduction.[6-8] We have recently proven the therapeutic effects of increased PA with or without dietary restriction in greatly reducing hepatic inflammation and the related oxidative stress levels outweigh those achieved by dietary restriction alone.[9,10] Thus, although there is an overall paucity of evidence on the benefits of PA as a treatment for NAFLD, management should include assessing PA levels and setting of lifestyle goals based on adopting regular exercise, with a focus on attaining sustainable PA habits.

Diet and PA interventions are important for NAFLD management, and there is increasing evidence that exercise beneficially modulates liver fat content. However, at present, clear guidelines for such a "lifestyle PA" for NAFLD management are lacking, and the dose (i.e., intensity and volume) of PA required to reduce liver fat content remains unclear.[11]

Considering this issue, we conducted a retrospective analysis of a large number of obese, middle-aged men with NAFLD who completed a 12-week supervised exercise plus dietary restriction program to determine the benefits of a varying PA dose (intensity and volume) in lifestyle modification on improving the pathophysiology of NAFLD.

Materials and Methods

Subjects. Figure 1 depicts the workflow of enrollment to the program, which was carried out in 2009 and 2011-2013 at the University of Tsukuba (Tsukuba, Japan). A total of 218 obese, middle-aged men (body mass index [BMI] 25-40 kg/m^2)[12] were recruited from Ibaraki prefecture through advertisements of weight reduction by means of a lifestyle management program by way of dietary restriction and exercise. The diagnosis of NAFLD was based on the diagnostic guidelines for NAFLD in the Asia-Pacific region.[13] For a comparative and thorough analysis, we excluded participants who withdrew, those without NAFLD, those <35 or >65 years old, those with BMI >40 kg/m^2, and those with an alcohol consumption of >21 units·wk^{-1}. Finally, of the initial 218 applicants, 169 subjects were enrolled and their data were analyzed for this retrospective study. Participants who performed moderate to vigorous intensity PA (MVPA) per week at PA level 4-9, estimated by using a uniaxial accelerometer, were classified into three groups: I_M group, <150 min·wk^{-1} (mean 101.6 ± 15.8 min·wk^{-1}, n = 40); II_M group, 150-250 min·wk^{-1} (mean 216.0 ± 15.4 min·wk^{-1}, n = 42); and III_M group, ≥250 min·wk^{-1} (mean 409.7 ± 10.7 min·wk^{-1}, n = 87) (for details, see Table 1). For further analysis,

Address reprint requests to: Junichi Shoda, M.D., Ph.D., Faculty of Medicine, University of Tsukuba, 1-1-1 Tennodai, Tsukuba, Ibaraki 305-8575, Japan. E-mail: shodaj@md.tsukuba.ac.jp; fax: +81 29 853 5795.

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we created two subgroups by using MVPA for 250 min·wk^{-1} as the cutoff (I_{M250} group: <250 min·wk^{-1} [mean 160.3 ± 7.20 min·wk^{-1}, n = 82]; II_{M250} group: ≥250 min·wk^{-1} [mean 409.7 ± 10.7 min·wk^{-1}, n = 87]). This allowed us to obtain more concrete data on the effects of a larger volume of MVPA on the risk factors for fibrosis progression and the changes of total fatty composition and related genes. The study protocol was approved by the Institutional Review Board of the University of Tsukuba. All participants provided written informed consent before their participation in the study.

Diet Restriction and Exercise Programs. The subjects were provided with a dietary program restricting their dietary intake to ∼1,680 kcal·d^{-1}. The dietary intake per meal (three times per day) was 80 kcal from eggs and/or dairy products; 80 kcal from vegetables and fruits; 160 kcal from meat, fish, and/or soybean products; and 240 kcal from carbohydrates and oils. During the 12 weekly lectures, the subjects mainly learned methods for calculating dietary calories by weighing food and planned a dietary program for maintaining 1,680 kcal·d^{-1}. After each lecture, dietitians checked the subjects' food diaries (on which daily dietary calories were recorded) and provided face-to-face dietary behavioral counseling.

The subjects also underwent an aerobic exercise program of 90 min·d^{-1}, 3 days·wk^{-1}. This program consisted of 40-60 minutes of walking and/or light jogging sessions and 15-25 minutes each of warm-up and cool-down sessions. Although they were given a few recommendations targeted to the Borg[14] scale ranging from 11 (light) to 13 (fairly hard), the subjects were free to determine the intensity and volume of exercise appropriate for their health condition. By taking part in the above sessions, the subjects learned about exercise methods, such as how to increase the exercise intensity, volume, and frequency according to their physical condition. On the basis of this education, the subjects were advised to keep performing aerobic exercise at or around their homes on days.

Energy Intake and PA Analysis. At baseline and at week 12, the study subjects maintained daily food-intake records for 3 consecutive days. A dietician used the food-intake records to estimate the total daily energy intake (TEI) and macronutrient composition (carbohydrate, protein, and fat intake), by using Excel Eiyo-Kun v. 4 software (Kenpakusya, Tokyo, Japan).

Daily PA monitoring in the aerobic exercise program and in free-living conditions was conducted using a uniaxial accelerometer (Lifecorder; Suzuken, Nagoya, Japan). As this device continuously measures the intensity, duration, and frequency of PA, it is useful for obtaining objective data on PA patterns and for estimating the related energy expenditure. Such PA information taken in 4-second sampling intervals was categorized into one of nine PA levels (1.0-9.0). Each PA level can be classified into a category of metabolic equivalents (METs) according to Kumahara et al.[15] PA levels estimated according to METs were also categorized on the basis of the intensity cutoff by Pate et al.,[16] and a PA level from 4.0 to 9.0 was used as the MVPA (for details, see Table 1).

Hepatic Stiffness and Steatosis. During 2011-2013, a clinical gastroenterologist assessed hepatic stiffness by using a Fibroscan device (Echosens, Paris, France) with the 3.5-MHz standard probe. The principles and examination procedures for such an assessment have been previously published.[17] In addition, the hepatic steatosis levels in 2012 and 2013 were determined by using a controlled attenuation parameter (CAP) designed to measure the liver ultrasonic attenuation at 3.5 MHz by using signals acquired with Fibroscan. Detailed descriptions of the CAP have also been previously published.[18]

Anthropometric Parameters. The body weight was measured with a digital electronic scale (TBF-551; Tanita, Tokyo, Japan). Their standing height was measured with a wall-mounted stadiometer (YG-200; Yagami, Nagoya, Japan). These data were used to calculate the BMI (kg/m^2). Total fat (TF) was assessed by using dual-energy x-ray absorptiometry (QDR 4500; Hologic, Bedford, MA). Abdominal adipose tissue was determined by using magnetic resonance imaging (3.0-Tesla system, Achieva R3.2; Philips, Best, the Netherlands) in which the visceral adipose tissue (VAT) area and subcutaneous adipose tissue (SAT) area were measured at the umbilicus level. Waist circumference (WC) was measured by using a fiberglass tape at the umbilicus level.

Blood Test. The levels of high-density lipoprotein-cholesterol (HDL-C) and triglycerides (TG) were analyzed enzymatically as follows: fasting plasma glucose level with a hexokinase-G-6-PDH method; fasting plasma insulin level with a chemiluminescent immunoassay method; aspartate aminotransferase (AST), alanine aminotransferase (ALT), and gamma glutamyl transpeptidase (γGT) levels with the Japan Society of Clinical Chemistry transferable method; hyaluronic acid (HA) and ferritin levels with the latex agglutination method; and high-sensitivity C-reactive protein (hs-CRP) level with a fixed time assay method. The platelets were counted on an automated analyzer (XE-2100; Sysmex, Kobe, Japan). We used these data to calculate the surrogate marker levels for insulin resistance by homeostasis model assessment (HOMA-IR) according to Matthews et al.[19] and for NAFLD fibrosis scores (NFS) according to the equation of Angulo et al.[20]

Table 1. PA Data (Intensity and Volume) Recorded With the Accelerometer for the 12-Week Weight Reduction Program in a Total of 169 Obese, Middle-Aged Men With NAFLD, With Stratification According to the Time of Moderate to Vigorous Intensity PA

		I_M	II_M	III_M	I_M vs. II_M	I_M vs. III_M	II_M vs. III_M
PA intensity*	**PA level [METs]†**	**N = 40**	**N = 42**	**N = 87**		***P* Value**	
Light intensity, $min \cdot wk^{-1}$	1.0 [1.8]	87.2 ± 6.61	100.5 ± 6.46	100.8 ± 4.49	0.327	0.211	0.999
	2.0 [2.3]	189.8 ± 13.0	215.5 ± 12.7	216.9 ± 8.82	0.338	0.200	0.995
	3.0 [2.9]	90.2 ± 7.15	108.6 ± 6.98	115.6 ± 4.85	0.158	<0.05	0.691
Moderate intensity, $min \cdot wk^{-1}$	4.0 [3.6]	61.1 ± 7.77	100.8 ± 7.58	156.1 ± 5.27	<0.01	<0.01	<0.01
	5.0 [4.3]	25.6 ± 9.03	62.5 ± 8.81	122.3 ± 6.12	<0.01	<0.01	<0.01
	6.0 [5.2]	10.9 ± 7.52	30.3 ± 7.33	78.7 ± 5.01	0.158	<0.01	<0.01
Vigorous intensity, $min \cdot wk^{-1}$	7.0 [6.1]	2.55 ± 3.31	8.16 ± 3.23	20.4 ± 2.25	0.385	<0.01	<0.01
	8.0 [7.1]	1.20 ± 3.42	11.8 ± 3.03	24.8 ± 2.32	<0.05	<0.01	<0.01
	9.0 [>8.3]	0.26 ± 1.82	2.64 ± 1.78	7.35 ± 1.24	0.622	<0.05	0.078
LPA (PA level 1.0-3.0), $min \cdot wk^{-1}$		367.2 ± 23.3	424.6 ± 22.8	433.3 ± 15.8	0.187	0.061	0.947
MVPA (PA level 4.0-9.0), $min \cdot wk^{-1}$		101.6 ± 15.8	216.0 ± 15.4	409.7 ± 10.7	<0.01	<0.01	<0.01
Energy expenditure, $kcal \cdot d^{-1}$		2388.3 ± 42.5	2437.1 ± 41.0	2576.1 ± 28.8	0.205	<0.01	<0.01
Step frequency, $step \cdot d^{-1}$		7367.1 ± 382.0	9075.5 ± 372.8	11779.4 ± 259.7	<0.01	<0.01	<0.01

Values are presented as the group means ± SE. To compare between groups, all dependent variables in PA levels were analyzed by using one-way ANOVA. *PA levels estimated by using METs were categorized according to the intensity cutoff of Pate et al.[16] (1.0 [PA level] = 1.8 [MET], 2.0 = 2.3, 3.0 = 2.9, 4.0 = 3.6, 5.0 = 4.3, 6.0 = 5.2, 7.0 = 6.1, 8.0 = 7.1, 9.0 = >8.3). †Each PA level measured with the accelerometer was classified into the category of METs according to Kumahara et al.[15] (light: <3.0 [MET], moderate: 3.0-6.0 and vigorous: >6.0).

PA, physical activity; METs, metabolic equivalents; LPA, light intensity PA; MVPA, moderate to vigorous intensity PA; I_M, the group who performed moderate to vigorous intensity PA for <150 $min \cdot wk^{-1}$; II_M, the group who performed moderate to vigorous intensity PA for 150-250 $min \cdot wk^{-1}$; III_M, the group who performed moderate to vigorous intensity PA for ≥250 $min \cdot wk^{-1}$.

Serum fatty acid compositions were analyzed with gas chromatography-mass spectrometry. Commercial enzyme-linked immunosorbent assay kits were used to measure the serum levels of thiobarbituric acid reactive substances (TBARS; Cayman Chemical, Ann Arbor, MI); tumor necrosis factor alpha (TNF-α), interleukin 6 (IL-6), and leptin (R&D Systems, Minneapolis, MN); M30 and M65 (Peviva; Bromma, Sweden); WFA^+-Mac-2 binding protein (WFA^+-M2BP; Sysmex, Kobe, Japan); M2BP (Immuno-Biological Lab, Kunma, Japan), and adiponectin (Sekisui Medical, Tokyo, Japan).

PBMC Isolation, RNA Isolation, and Reverse-Transcription, Real-Time Quantitative Polymerase Chain Reaction (PCR). The gene expression levels in peripheral blood mononuclear cells (PBMCs) can reflect those in the liver.[21] PBMCs are considered a good model to reflect important metabolic changes in the liver.[22]

PBMCs were isolated from LSM density gradients (MP Biomedical, Santa Ana, CA). From cell pellets containing the PBMCs, RNA was extracted. Subsequently, first-strand complementary DNA was synthesized by using a PrimeScript RT reagent kit (Takara Bio, Shiga, Japan). The complementary DNA templates were added to Fast SYBR Green Master Mix (Applied Biosystems, Santa Ana, CA). PCR was performed on a CFX384 Touch Real-Time PCR Detection System (Bio-Rad Laboratories, Foster City, CA). The primers (FASMAC, Tokyo, Japan) are shown in Supporting Material 1.

Statistics. Statistical analysis was performed using SPSS 20.0 (IBM, Armonk, NY). Descriptive parameters were given as mean ± SE or log transformations for skewed variables, and as percentages for categorical variables. To compare groups for all dependent variables at baseline and PA data, we performed one-way analysis of variance (ANOVA). Categorical variables, the chi-square test, or Fisher's exact test was used. To compare intra- and intergroup changes over time (at baseline and the 12th week), all dependent variables of the pathophysiological factors of NAFLD were subjected to a repeated-measure ANOVA with/without the change in weight as a covariate. $P < 0.05$ was considered significant.

Results

Baseline Characteristics. There were no significant differences in age, BMI, PA, alcohol intake, smoking, or medication use of subjects among the three groups (Table 2). The mean values of all parameters including dietary intake (Table 3), body adiposity (Table 4), and blood test (Table 4) were not significantly different between the three groups. The attendance rate was 81.2% in the I_M group, 80.3% in the II_M group, and 85.3% in the III_M group. The difference was not statistically significant.

PA Data. Table 1 shows the results of PA recorded with the accelerometer during the

Table 2. Baseline Characteristics of a Total of 169 Obese, Middle-Aged Men With NAFLD Who Participated in a Weight Reduction Program, With Stratification According to the Time of Moderate to Vigorous Intensity PA

	I_M	II_M	III_M	
Parameters	**Baseline**	**Baseline**	**Baseline**	***P* Value**
n	40	42	87	
Age, *year*	52.6 ± 1.30	49.0 ± 1.27	51.9 ± 0.88	0.096
BMI, kg/m^2	29.4 ± 0.51	28.8 ± 0.50	29.2 ± 0.35	0.673
Physical activity				
Energy expenditure, $kcal \cdot d^{-1}$	2354.3 ± 40.1	2351.4 ± 40.0	2425.3 ± 27.8	0.182
Step frequency, $steps \cdot d^{-1}$	7277.9 ± 312.4	7497.7 ± 304.9	7944.9 ± 204.8	0.151
Alcohol intake, $unit \cdot wk^{-1}$	7.68 ± 1.46	8.92 ± 1.34	7.92 ± 1.04	0.785
Smoking, *% subject*	15.0	19.0	17.2	0.888
Medication				
Hyperglycemic agents, *% subject*	7.5	2.4	5.7	0.569
Hypertensive agents, *% subject*	32.5	26.2	32.2	0.759
Hyperlipidemia agents, *% subject*	15.0	19.0	18.4	0.869

Values are presented as the group means ± SE and as percentages. To compare between groups, all dependent variables at baseline were analyzed by using one-way ANOVA. In the case of categorical variables, the chi-square test or Fisher's exact test was used.

BMI, body mass index; I_M, the group who performed moderate to vigorous intensity PA for <150 $min \cdot wk^{-1}$; II_M, the group who performed moderate to vigorous intensity PA for 150-250 $min \cdot wk^{-1}$; III_M, the group who performed moderate to vigorous intensity PA for ≥ 250 $min \cdot wk^{-1}$.

intervention periods. When a comparison was made between the groups, the mean values of the energy expenditure and step frequency were greater in the III_M group than those in the I_M and II_M groups ($P < 0.01$). A comparison between I_M and II_M revealed that the magnitude of the change in step frequency was greater in the II_M group ($P < 0.01$) than in the I_M group. The mean value of the light-intensity PA was not significantly different between the three groups, whereas there was a statistical difference in the MVPA between each group ($P < 0.01$).

Dietary Intake. Table 3 shows the outcomes of dietary intake during the intervention period. According to the daily food-intake records, all groups had significantly reduced TEI and carbohydrate, protein, and fat intake between baseline and week 12 ($P < 0.01$). When an analysis with group-by-time interactions was made the magnitude of change in all outcomes showed no significant differences between the groups.

Body Weight. Weight and BMI (Table 4) were significantly reduced at week 12 in each group ($P < 0.01$). When comparing group-by-time interaction between groups, these parameters were significantly reduced for the II_M and III_M groups compared with the I_M group ($P < 0.01$). However, a comparison between II_M and III_M found no statistically significant difference.

Body Adiposity. The results of the evaluations of body composition and abdominal distribution (Table 4) revealed that all parameters were improved at week 12 in each group ($P < 0.01$). When comparing group-by-time interactions between groups with adjustment for change in weight, all body adiposity parameters significantly improved in the II_M and III_M groups compared with those in the I_M group ($P < 0.01$), except for the SAT area. A comparison between II_M and III_M with adjustment for change in weight revealed that the magnitude of change in the VAT area was greater in the III_M group ($P < 0.05$) than in the II_M group.

Blood Test. Among the 13 parameters in blood analysis (Table 4), eight parameters ($_{log}$TG, HOMA-IR, AST, ALT, γGT, $_{log}$hs-CRP, $_{log}$leptin, and $_{log}$TNF-α) in the I_M group ($P < 0.05$), nine parameters ($_{log}$TG, HDL-C, HOMA-IR, AST, ALT, γGT, $_{log}$hs-CRP, $_{log}$leptin, and $_{log}$TNF-α) in the II_M group ($P < 0.05$), and all 13 parameters in the III_M group ($P < 0.01$) improved at week 12. Parameters that did not show improvement in the I_M group were the serum levels of HDL-C, ferritin, TBARS, adiponectin, and IL-6, whereas those who failed to show improvement in the II_M group were the serum levels of ferritin, TBARS, adiponectin, and IL-6.

Analysis of group-by-time interactions with adjustment for change in weight revealed that the magnitude of change in the serum levels of $_{log}$TG was greater in the II_M group than in the I_M group ($P < 0.05$), and the magnitude of change in the serum levels of $_{log}$TG, HDL-C, ferritin, TBARS, and adiponectin was greater in the III_M group than in the I_M group ($P < 0.05$). A comparison between the II_M and III_M groups with the change in weight as a covariate revealed that only four parameters reflecting the magnitude of the increase (HDL-C and adiponectin) and the decrease (ferritin and TBARS) were greater in the III_M group than in the II_M group ($P < 0.05$).

Apoptosis and Fibrosis Biomarkers. Six molecules were selected as hepatocyte apoptosis and liver fibrosis biomarkers (apoptosis: M30, M65; fibrosis: M2BP, WFA^+-

Table 3. Outcomes of Body Weight and Dietary Intake of a Total of 169 Obese, Middle-Aged Men With NAFLD, With Stratification According to the Time of Moderate to Vigorous Intensity PA

	I_M (n = 40)			II_M (n = 42)			III_M (n = 87)					
	Mean: 101.6 min·wk^{-1}/ 14.5 min·d^{-1}			Mean: 216.0 min·wk^{-1}/ 30.9 min·d^{-1}			Mean: 409.7 min·wk^{-1}/ 58.5 min·d^{-1}			I_M vs. II_M	I_M vs. III_M	II_M vs. III_M
										Time × Group Interaction *P* Value		
Parameter	Baseline	After	Change	Baseline	After	Change	Baseline	After	Change			
Weight, *kg*	86.1 ± 2.15	80.6 ± 1.84	−5.5†	82.9 ± 1.39	73.0 ± 1.24	−9.9†	84.3 ± 1.18	73.4 ± 1.11	−10.9†	<0.01	<0.01	0.209
BMI, *kg/m*^{2}	29.4 ± 0.51	27.5 ± 0.48	−1.9†	28.8 ± 0.50	25.3 ± 0.47	−3.4†	29.2 ± 0.35	25.5 ± 0.33	−3.7†	<0.01	<0.01	0.227
TEI, *kcal·d*$^{-1}$	2199.1 ± 75.2	1542.2 ± 43.1	−656.8†	2177.5 ± 70.2	1490.4 ± 40.7	−687.2†	2231.0 ± 51.2	1545.5 ± 29.5	−685.6†	0.581	0.773	0.412
Carbohydrate, *g·d*$^{-1}$	295.9 ± 12.1	216.3 ± 7.05	−79.6†	278.9 ± 11.0	199.6 ± 6.42	−79.2†	299.4 ± 8.21	208.5 ± 4.56	−90.9†	0.981	0.433	0.114
Protein, *g·d*$^{-1}$	78.7 ± 3.04	68.8 ± 2.27	−9.9†	77.4 ± 3.12	65.4 ± 2.18	−12.0†	80.9 ± 2.07	72.8 ± 1.55	−8.1†	0.618	0.595	0.301
Fat, *g·d*$^{-1}$	60.5 ± 2.71	40.5 ± 1.79	−20.0†	65.4 ± 2.81	43.1 ± 1.74	−22.3†	62.5 ± 1.86	43.0 ± 1.21	−19.5†	0.602	0.864	0.902

Values are presented as the group means ± SE. To compare intergroup changes over time, from baseline to 12 weeks, all dependent variables were analyzed by using a repeated ANOVA. †$P < 0.01$, significant difference within group.

BMI, body mass index; PA, physical activity; TEI, total daily energy intake. I_M, the group who performed moderate to vigorous intensity PA for <150 min·wk^{-1}; II_M, the group who performed moderate to vigorous intensity PA for 150-250 min·wk^{-1}; III_M, the group who performed moderate to vigorous intensity PA for ≥250 min·wk^{-1}.

M2BP, HA, and NFS). Figure 2A shows the results of these parameter changes in each group from baseline to week 12. All biomarkers except NFS ($P = 0.12$) for the 82 subjects in the I_{M250} group and the 87 subjects in the II_{M250} group were decreased at week 12 ($P < 0.05$). When an analysis of group-by-time interactions with adjustment for change in weight was made between the groups, no statistically significant difference was found.

Hepatic Stiffness and Steatosis. The subjects in the 2012 and 2013 interventions were evaluated for hepatic steatosis levels by using CAP (Fig. 2B). In addition, hepatic stiffness was assessed between 2011 and 2013. From the CAP, a significant reduction in hepatic steatosis levels was seen after the 12-week program in the 31 subjects in the I_{M250} group and in the 49 subjects in the II_{M250} MVPA group (−23.2% and −31.8%, respectively). When a group-by-time interaction analysis with adjustment for change in weight was performed and compared between the groups, the magnitude of the changes in hepatic steatosis was greater in the II_{M250} group than in the I_{M250} group ($P < 0.05$). Hepatic stiffness also significantly improved in both groups (I_{M250} n = 41, II_{M250} n = 69) from baseline to week 12 (−21.0% and −21.9%, respectively). However, a comparison between these groups with the change in weight as a covariate revealed that the magnitude of the changes in stiffness was not significantly different ($P = 0.64$).

Fatty Acid Composition. The serum fatty acid compositions recorded between 2011 and 2013 are shown in Table 5. The total fatty acid parameters for the 41 subjects in the I_{M250} group and the 69 subjects in the II_{M250} group were significantly decreased at week 12 (I_{M250}: −19.8%, II_{M250}: −26.9%; $P < 0.01$). The total saturated fatty acid, total monounsaturated fatty acid, and total polyunsaturated fatty acid levels also significantly decreased in both groups ($P < 0.05$). When an analysis of group-by-time interactions was performed and compared between the groups with the change in weight as a covariate, the magnitude of change in all outcomes was not significantly different.

Expression Levels of Genes Involved in Fatty Acid Synthesis and Degradation. Table 5 shows the changes in the expression levels in the PBMCs of theeight genes involved in fatty acid synthesis ($_{log}$SREBP1c [sterol regulatory element-binding protein 1c], $_{log}$FAS [fatty acid synthase], $_{log}$SCD1 [stearoyl-CoA desaturase-1], $_{log}$ACC [acetyl-CoA carboxylase], $_{log}$EVLOVE6) and degradation ($_{log}$ACS [acyl-CoA synthetase], $_{log}$CPT1 [carnitine palmitoyltransferase I], $_{log}$acyl-CoA oxidase) in each group between baseline and week 12. Among the eight PBMC genes, three ($_{log}$FAS, $_{log}$ACC, $_{log}$ACS)in the I_{250M} group ($P < 0.05$) and seven

Table 4. Parameter Outcomes of Body Adiposity, Lipid Profiles, and Insulin Resistance, Liver Function Test, Inflammation and Oxidative Stress, and Adipocytokine Values in a Total of 169 Obese, Middle-Aged Men With NAFLD, With Stratification According to the Time of Moderate to Vigorous Intensity PA

	I_M (n = 40)			II_M (n = 42)			III_M (n = 87)			I_M vs. II_M	I_M vs. III_M	II_M vs. III_M
	Mean: 101.6 min·wk^{-1}/14.5 min·d^{-1}			Mean: 216.0 min·wk^{-1}/30.9 min·d^{-1}			Mean: 409.7 min·wk^{-1}/58.5 min·d^{-1}			Time × Group Interaction *P* Value		
Parameter	Baseline	After	Change	Baseline	After	Change	Baseline	After	Change			
Body adiposity												
WC, *cm*	101.7 ± 1.77	96.4 ± 1.62	−5.3†	98.9 ± 0.95	88.5 ± 1.00	−10.4†	99.8 ± 0.76	88.7 ± 0.95	−11.0†	< 0.01	< 0.05	0.694
TF, *%*	25.3 ± 0.65	23.3 ± 0.61	−2.1†	25.3 ± 0.49	21.0 ± 0.56	−4.3†	24.3 ± 0.37	19.4 ± 0.43	−4.9†	< 0.01	< 0.01	0.334
VAT area, cm^2	163.2 ± 8.06	142.2 ± 7.33	−21.0†	149.9 ± 4.89	105.5 ± 5.13	−44.4†	166.4 ± 6.68	98.8 ± 5.26	−67.6†	< 0.05	< 0.01	< 0.01
SAT area, cm^2	223.7 ± 16.1	181.1 ± 12.2	−42.6†	230.9 ± 10.1	153.9 ± 7.57	−77.0†	229.0 ± 7.37	150.2 ± 6.99	−78.8†	0.400	0.603	0.329
Lipid profiles and insulin resistance												
$_{log}$TG	2.12 ± 0.03	2.02 ± 0.03	−0.10†	2.14 ± 0.04	1.87 ± 0.03	−0.27†	2.13 ± 0.03	1.85 ± 0.02	−0.28†	< 0.05	< 0.01	0.983
HDLC, *mg/dL*	46.5 ± 1.51	47.3 ± 1.50	+0.8	47.9 ± 1.66	50.1 ± 1.66	+2.2*	50.9 ± 1.04	56.1 ± 1.13	+5.2†	0.121	< 0.05	0.069
HOMA-IR	2.61 ± 0.22	2.03 ± 0.26	−0.58*	2.64 ± 0.37	1.25 ± 0.13	−1.39†	2.39 ± 0.21	1.26 ± 0.22	−1.13†	0.595	0.412	0.256
Liver function test												
AST, *U/L*	24.8 ± 1.44	21.5 ± 0.73	−3.2†	25.5 ± 1.46	20.8 ± 0.94	−4.7†	25.9 ± 1.19	21.4 ± 0.82	−4.5†	0.894	0.294	0.630
ALT, *U/L*	34.3 ± 2.89	26.1 ± 1.70	−8.2†	36.3 ± 3.67	22.9 ± 1.58	−13.5†	33.7 ± 2.70	22.0 ± 1.18	−11.7†	0.563	0.432	0.414
γGT, *U/L*	61.6 ± 8.61	43.1 ± 4.88	−18.5†	61.9 ± 9.16	29.1 ± 2.96	−32.8†	49.4 ± 2.98	25.3 ± 1.61	−24.1†	0.440	0.892	0.095
Inflammation and oxidative stress												
$_{log}$ hs-CRP	1.81 ± 0.08	1.67 ± 0.08	−0.14*	1.78 ± 0.07	1.59 ± 0.07	−0.19†	1.81 ± 0.05	1.56 ± 0.05	−0.25†	0.489	0.155	0.315
Ferritin, μg*/L*	225.7 ± 16.2	229.1 ± 17.4	+3.4	203.9 ± 15.9	195.9 ± 16.9	−8.0	197.9 ± 11.0	171.0 ± 11.8	−26.9†	0.209	< 0.05	< 0.05
TBARS, μ*M/L*	21.7 ± 1.28	21.1 ± 1.10	−0.6	23.4 ± 1.41	22.2 ± 1.41	−1.2	22.5 ± 0.97	19.1 ± 0.66	−3.4†	0.814	< 0.01	< 0.01
Adipocytokines												
Adiponectin, μg*/mL*	3.93 ± 0.27	4.05 ± 0.32	+0.22	4.13 ± 0.33	4.36 ± 0.32	+0.23	4.85 ± 0.25	5.68 ± 0.24	+0.83†	0.459	< 0.05	< 0.05
$_{log}$ Leptin	4.73 ± 0.04	4.48 ± 0.04	−0.25†	4.82 ± 0.04	4.46 ± 0.05	−0.36†	4.82 ± 0.26	4.43 ± 0.33	−0.39†	0.214	0.324	0.890
$_{log}$TNF-α	1.36 ± 0.04	1.94 ± 0.08	−0.42†	1.32 ± 0.04	0.99 ± 0.07	−0.33†	1.34 ± 0.03	1.12 ± 0.04	−0.22†	0.193	0.062	0.087
IL-6, *pg/mL*	1.38 ± 0.13	1.22 ± 0.13	−0.16	1.46 ± 0.15	1.21 ± 0.17	−0.25	1.65 ± 0.12	1.26 ± 0.09	−0.39†	0.865	0.099	0.371

Values are presented as the group means ± SE. To compare intergroup changes over time, from baseline to 12 weeks, all dependent variables were analyzed by using a repeated ANOVA with the change in weight as a covariate. †$P < 0.01$ and *$P < 0.05$, significant difference within group.

PA, physical activity; WC, waist circumference; TF, total fat; VAFA, visceral abdominal fat area; SAFA, subcutaneous abdominal fat area; TG, triglyceride; HDLC, high density lipoprotein cholesterol; HOMA-IR, insulin resistance by homeostasis model assessment; AST, aspartate aminotransferase; ALT, alanine aminotransferase; γGT, gamma glutamyl transpeptidase; hs-CRP, high-sensitivity C-reactive protein; TBARS, thiobarbituric acid reactive substances; TNF-α, tumor necrosis factor alpha; IL-6, interleukin 6; I_M, the group who performed moderate to vigorous intensity PA for <150 min·wk^{-1}; II_M, the group who performed moderate to vigorous intensity PA for 150-250 min·wk^{-1}; III_M, the group who performed moderate to vigorous intensity PA for ≥250 min·wk^{-1}.

(except $_{log}$SCD1) in the II_{250M} group ($P < 0.05$) were found to be significantly different. When a comparison was made between the two groups with the change in weight as a covariate, only two genes showed a magnitude of decrease ($_{log}$SREBP1c) or increase ($_{log}$CPT1) that was greater in the III_M group than in the II_M group ($P < 0.05$).

Discussion

The major findings of this study are that MVPA for ≥250 min·wk^{-1} in comparison with MVPA for <250 min·wk^{-1} attenuates the degree of hepatic steatosis independent of weight reduction. In relation, MVPA for ≥250 min·wk^{-1} induced a significant improvement in the VAT severity and levels of ferritin, lipid peroxidation, and adiponectin (Fig. 3A), as well as altered fatty acid metabolism. Moreover, MVPA for 150-250 min·wk^{-1}, in comparison with MVPA for <150 min·wk^{-1}, also induced a significant improvement in the WC, TF, and VAT severity and TG level (Fig. 3B,C). Overall, the results suggest that an increase in MVPA per week benefits the management of NAFLD. To the best of our knowledge, these results provide the first clinical evidence of the benefits of an increase in MVPA, which, in turn, is used as a "lifestyle PA" in NAFLD management.

In this study, MVPA for >150 min·wk^{-1} achieved a 12.4% reduction in weight for obese, middle-aged men with NAFLD, whereas MVPA for <150 min·wk^{-1} showed a 6.4% reduction in the equivalent amount of calorie intake. This observed weight-reduction-effect of MVPA for >150 min·wk^{-1} is important for NAFLD management. Moreover, to investigate the significance of MVPA volume regardless of weight reduction on NAFLD risk factors, all parameters were analyzed with/without the change in weight as a covariate. After controlling for change in weight, the magnitude of MVPA for ≥250 min·wk^{-1} on insulin resistance and inflammatory adipokines (leptin and TNF-α) became

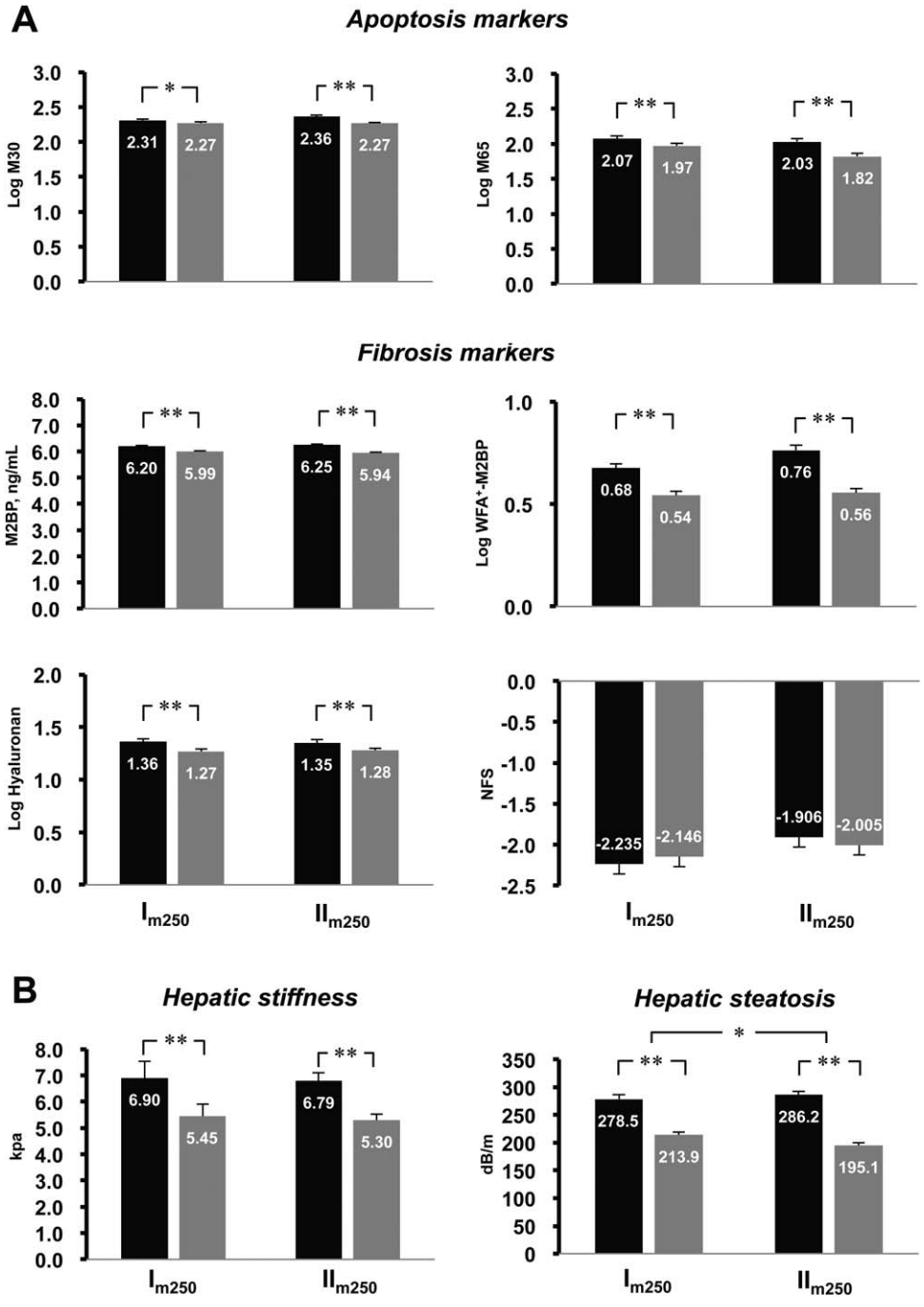

Fig. 2. Changes in the levels of apoptosis and fibrosis markers (total number of subjects = 169; I_{M250} = 76, II_{M250} = 87) (A) and the degree of hepatic steatosis (total, 80; I_{M250} = 31, II_{M250} = 49) and stiffness (total, 110; I_{M250} = 41, II_{M250} = 69) (B) from baseline to week 12 in obese, middle-aged men. Values are presented as the group means ± SE. A repeated ANOVA model with the change in weight as a covariate was used to compare the intra- and inter-group changes over time, from baseline to week 12. The black bar indicates baseline and the gray bars indicate after week 12 (means ± SE). $^{**}P < 0.01$, $^{*}P < 0.05$, significantly different between baseline and week 12; brackets $^{**}P < 0.01$, $^{*}P < 0.05$, significantly different MVPA performed per week between the groups.

statistically nonsignificant; however, that in the change in hepatic steatosis, VAT, TG, HDLC, ferritin, TBARS, and adiponectin remained significant. Collectively, the effects of MVPA seem to be associated with an improvement in the hepatic inflammatory conditions and related oxidative stress levels. These benefits might be independent of weight reduction.

In subjects with NAFLD, the clinical benefits observed in those who performed MVPA for ≥250 min·wk^{-1} are the improved inflammatory and oxidative stress levels, reflected by higher serum ferritin and TBARS levels.[23] Hepatic iron overload, reflected by elevated serum ferritin levels[24] and dysregulation of adipokine production, further increases the intrahepatic oxidative stress levels.[25] This, in turn, causes impaired nucleotide and protein synthesis, leading to apoptosis, inflammation, and liver fibrosis.[25] Therefore, the potent reduction in hepatic fat content and serum ferritin and TBARS levels in subjects who performed MVPA for ≥250 min·wk^{-1} may have contributed to improved NAFLD pathophysiology. The results indicated the advantage of adding MVPA for

Table 5. Serum Total Fatty Acid Composition and Expression Levels of Genes Involved in Fatty Acid Metabolism for a Total of 110 Obese, Middle-Aged Men With NAFLD, With Stratification According to the Time of Moderate to Vigorous Intensity PA

	I_{M250} (n = 41)			II_{M250} (n = 69)			I_{M250} vs. II_{M250}
Parameter	**Baseline**	**After**	**Change**	**Baseline**	**After**	**Change**	**Time x Group Interaction *P* Value**
Fatty acid composition (μg/mL)							
Total SFA	1387.5 ± 84.9	1092.1 ± 59.1	−295.4†	1311.6 ± 59.1	916.2 ± 27.2	−395.4*	0.556
C14:0	36.8 ± 4.67	25.6 ± 5.32	−11.2*	33.4 ± 3.18	11.9 ± 3.62	−21.5†	0.267
C16:0	1010.0 ± 63.3	803.5 ± 35.8	−206.5†	957.0 ± 43.1	680.4 ± 24.4	−276.6†	0.620
C18:0	299.0 ± 16.3	228.5 ± 10.0	−70.5†	280.3 ± 11.1	191.5 ± 6.83	−88.8†	0.857
C22:0	21.0 ± 0.70	17.8 ± 0.61	−3.2†	20.4 ± 0.47	16.6 ± 0.41	−3.8†	0.586
C24:0	20.7 ± 0.66	16.7 ± 0.54	−4.0†	20.5 ± 0.45	15.8 ± 0.37	−4.7†	0.598
Total MUFA	1028.2 ± 65.1	750.5 ± 32.9	−277.7†	925.3 ± 43.9	623.0 ± 20.2	−302.3†	0.978
C16:1 W-7	77.6 ± 7.33	43.2 ± 3.39	−34.4†	71.9 ± 5.00	32.5 ± 2.31	−39.5†	0.623
C18:1 W-9	906.8 ± 57.8	660.7 ± 28.7	−246.1†	809.6 ± 39.4	541.3 ± 19.6	−268.3†	0.982
C20:1 W-9	6.12 ± 0.56	4.21 ± 0.18	−1.9†	5.53 ± 0.38	3.93 ± 0.13	−1.6†	0.655
C24:1 W-9	37.7 ± 1.40	42.4 ± 1.67	+4.7†	38.3 ± 0.95	45.3 ± 1.14	+7.0†	0.939
Total PUFA	1526.1 ± 53.2	1320.3 ± 39.8	−205.8†	1529.4 ± 41.5	1212.9 ± 24.6	−316.5†	0.305
C18:2 W-6	1112.5 ± 44.4	940.2 ± 28.3	−172.3†	1081.5 ± 30.2	838.1 ± 19.3	−243.4†	0.541
C20:2 W-6	7.33 ± 0.44	4.91 ± 0.23	−2.42†	7.14 ± 0.30	4.56 ± 0.16	−2.58†	0.867
C20:4 W-6	202.9 ± 10.0	187.3 ± 6.90	−15.6	207.3 ± 6.83	181.0 ± 4.70	−26.3†	0.128
C20:5 W-3	60.4 ± 9.78	55.1 ± 7.05	−8.3	76.7 ± 6.66	61.1 ± 4.80	−15.6†	0.554
C22:4 W-6	5.33 ± 0.43	3.92 ± 0.26	−1.41†	4.87 ± 0.29	3.43 ± 0.18	−1.44†	0.686
C22:6 W-3	137.6 ± 12.4	128.9 ± 8.27	−8.7	151.9 ± 8.44	124.7 ± 5.63	−27.2†	0.123
Genes involved in fatty acid synthesis							
$_{log}$SREBP1c	2.51 ± 0.06	2.58 ± 0.07	+0.07	2.52 ± 0.05	2.37 ± 0.05	−0.15*	<0.05
$_{log}$FAS	2.31 ± 0.09	2.78 ± 0.10	+0.476*	2.28 ± 0.07	2.55 ± 0.08	+0.27*	0.533
$_{log}$SCD1	1.30 ± 0.13	1.39 ± 0.12	+0.09	1.23 ± 0.09	1.32 ± 0.09	+0.09	0.990
$_{log}$ACC	2.35 ± 0.04	2.19 ± 0.05	−0.16†	2.38 ± 0.03	2.25 ± 0.04	−0.13†	0.663
$_{log}$EVLOVE6	1.96 ± 0.06	1.94 ± 0.04	−0.02	2.02 ± 0.05	1.91 ± 0.03	−0.11*	0.222
Genes involved in fatty acid degradation							
$_{log}$ACS	2.34 ± 0.07	2.75 ± 0.06	+0.41†	2.36 ± 0.05	2.79 ± 0.05	+0.43†	0.994
$_{log}$CPT1	2.22 ± 0.05	2.15 ± 0.07	−0.07	2.08 ± 0.04	2.17 ± 0.05	+0.09*	<0.05
$_{log}$ACO	2.20 ± 0.04	2.29 ± 0.04	+0.10*	2.22 ± 0.03	2.32 ± 0.03	+0.10†	0.963

Values are presented as the group means ± SE. To compare intergroup changes over time, from baseline to 12 weeks, all dependent variables were analyzed by using a repeated ANOVA with the change in weight as a covariate. †$P < 0.01$ and *$P < 0.05$, significant difference within group.

PA, physical activity; C14:0, myristic acid; C16:0, palmitic acid; C18:0, stearic acid; C22:0, behenic acid; C24:0, lignoceric acid; C16:1 W-7, palmitoleic acid; C18:1 W-9, oleic acid; C20:1 W-9, eicosenoic acid; C24:1 W-9, nervonic acid; C18:2 W-6, linoleic acid; C20:2 W-6, eicosadienoic acid; C20:4 W-6, arachidonic acid; C20:5 W-3; eicosapentaenoic acid; C22:4 W-6, docosatetraenoic acid; C22:6 W-3, docosahexaenoic acid; SREBP1c, sterol regulatory element-binding protein 1c; FAS, fatty acid synthase; ACC, acetyl-CoA carboxylase; SCD1, stearoyl-CoA desaturase-1; ACS, acyl-CoA synthetase; CPT1, carnitine palmitoyltransferase I; ACO, acyl CoA oxidase; I_{M250}, the group who performed moderate to vigorous intensity PA for <250 min·wk^{-1}; II_{M250}, the group who performed moderate to vigorous intensity PA for ≥250 min·wk^{-1}.

≥250 min·wk^{-1} to dietary restriction. Because of the lack of histology data, the benefits of MVPA for ≥250 min·wk^{-1} in terms of antifibrosis effects could not be evaluated in this study. Nevertheless, the results of improvement in VAT, ferritin, TBARS, adiponectin, and other variables may indicate a potential improvement in inflammatory liver injuries associated with NASH. Recently, on the basis of liver histology, vigorous PA was related to a decrease in NASH risk found in a retrospective analysis of PA data from adult subjects with NAFLD.[26] Inflammation plays a central role in the onset of NASH.[27] Therefore, the antiinflammatory and/or antioxidative stress effects induced by MVPA for ≥250 min·wk^{-1} may be important in terms of the lifestyle management for NAFLD.

The details of such a mechanism underlying the observed clinical benefits are not fully understood; however, evidence from our laboratory and that of others may help clarify this issue. Exercise-induced oxidative stress, especially during high-intensity exercise such as MVPA for ≥250 min·wk^{-1}, triggers the activation of a redox-sensitive transcription factor known as nuclear factor E2-related factor 2 (Nrf2).[28] Nrf2 serves as an oxidative stress sensor and master regulator of the antioxidant response.[29] In this study, MVPA for ≥250 min·wk^{-1} significantly increased in obese subjects the expression levels of NADH quinone oxidoreductase (an enzyme involved in cellular detoxification) and ferropotin-1 (a transporter involved in cellular iron homeostasis), known as prototypical Nrf2 target genes (Supporting Material 2). Moreover, Nrf2 activation down-regulates the expression and activity of fatty acid synthesis enzymes,[30] which are associated with hepatic steatosis. Comparable to this, MVPA for ≥250

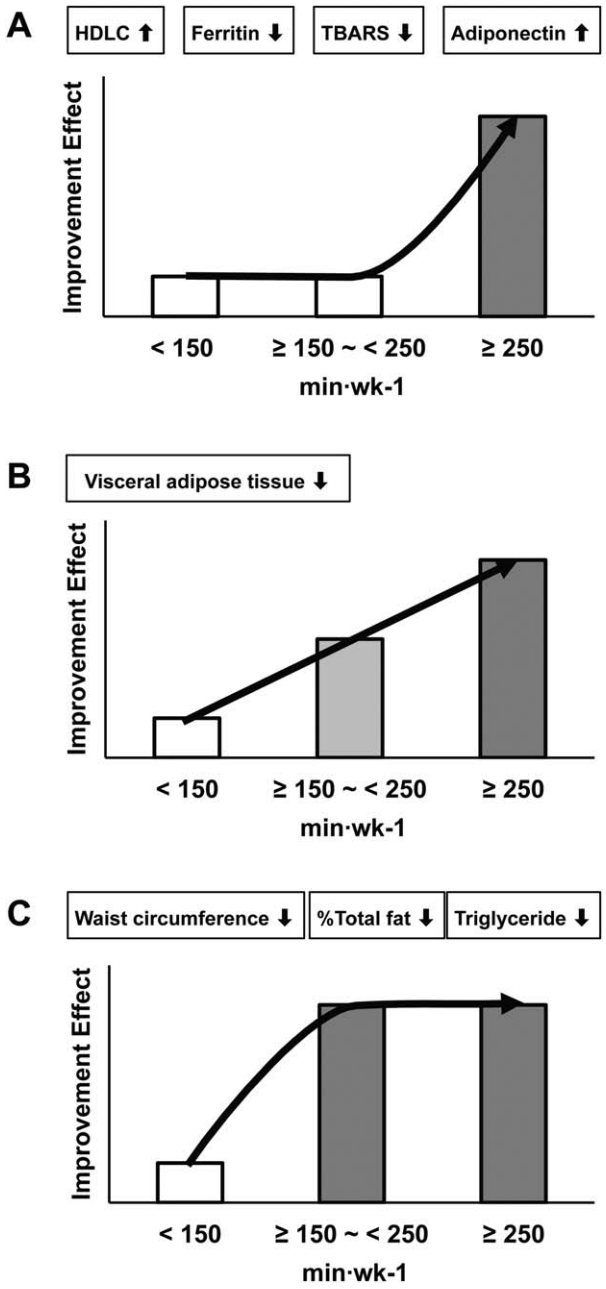

Fig. 3. Schematic summary of the beneficial effects of varying MVPA doses in lifestyle intervention on the pathophysiology of obesity-related liver disease in obese, middle-aged men. MVPA for $\geq$250 min·wk^{-1} combined with dietary restriction enhances the treatment effect by managing hepatic lipid levels through the modification of imbalanced adipokine levels and increased inflammation and oxidative stress levels in the liver (A). In addition, increasing the volume of MVPA contributes to improvements in obesity-related liver disease through reduction in the risk of abdominal obesity (B). Finally, MVPA for $\geq$150 min·wk^{-1} coupled with dietary restriction may arrest obesity and lipid profiles and thus also potentially improve obesity-related liver disease (C).

min·wk^{-1} decreased the SREBP1c expression involved in fatty acid synthesis and increased the CPT1 levels involved in fatty acid degradation (Table 5).

Adipokines, which are produced in adipose tissue, have a close relation with NAFLD pathology.[31] Therefore, it is noteworthy that MVPA for $\geq$250 min·wk^{-1} led to a marked increase in the adiponectin levels in comparison with MVPA for $<$150 min·wk^{-1} and MVPA for 150-250 min·wk^{-1} (Table 4). Thus, MVPA for $\geq$250 min·wk^{-1} is further beneficial as a lifestyle PA program in obese subjects. Low adiponectin levels are implicated in hepatic lipid accumulation and associated with NAFLD severity.[32] On the other hand, elevated adiponectin levels improve insulin sensitivity and enhance fatty acid oxidation, which in turn improve NAFLD pathology.[32] In addition, in one report, hepatic adiponectin signaling played a protective role against progression from simple steatosis to NASH in mice.[33] It is likely that the increased serum adiponectin level resulting from MVPA for $\geq$250 min·wk^{-1} contributes to the improved pathophysiology of NAFLD.

Concerning clinical relevance, increasing exercise frequency and dose, to achieve an MVPA of $\geq$250 min·wk^{-1}, is difficult for most obese subjects with NAFLD. In some obese subjects, continuous training with any type of exercise with less frequency and dose per week is recommended. The results of this study could address this critical issue. MVPA for 150-250 min·wk^{-1} yielded similar health benefits to those performing MVPA for $\geq$250 min·wk^{-1}, in terms of hepatic steatosis and the measured apoptosis and fibrosis markers. However, MVPA for $<$250 min·wk^{-1} did not affect HDL-C, ferritin, TBARS, adiponectin, VAT, SREBP1c, and CPT1 levels (Tables 3 and 4; Fig. 2B). Several studies have reported indications for obtaining the direct effect of exercise on hepatic steatosis[6-8]; exercise with less frequency and dose was found to be effective in both light and moderate volume exercise programs,[34] the PA volume of which was below the current guidelines for health promotion[35] and for managing body weight.[36] At present, scientific evidence for the benefits of PA and the optimal dose and modality for PA in NAFLD management is limited.[11]

In summary, the results of this study show the beneficial effects of increasing the volume of MVPA combined with dietary restriction on managing NAFLD in obese, middle-aged men. In the near future, prospective studies are needed to improve the objective criteria for the optimal intensity, duration, or total volume of MVPA; this is necessary to obtain beneficial effects in the management of NAFLD. Clarification of the criteria will enable the formulation of an effective and time-efficient PA program for improved outcomes, participation, and adherence of obese, middle-aged men.

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Author names in bold designate shared co-first authorship.

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