JOURNAL OF THE AMERICAN COLLEGE OF CARDIOLOGY VOL. 81, NO. 14, 2023

PUBLISHED BY ELSEVIER

Prolonged Moderate-Intensity Exercise Does Not Increase Muscle Injury Markers in Symptomatic or Asymptomatic Statin Users

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ABSTRACT

BACKGROUND Statin use may exacerbate exercise-induced skeletal muscle injury caused by reduced coenzyme Q10 (CoQ10) levels, which are postulated to produce mitochondrial dysfunction.

OBJECTIVES We determined the effect of prolonged moderate-intensity exercise on markers of muscle injury in statin users with and without statin-associated muscle symptoms. We also examined the association between leukocyte CoQ10 levels and muscle markers, muscle performance, and reported muscle symptoms.

METHODS Symptomatic (n = 35; age 62 ± 7 years) and asymptomatic statin users (n = 34; age 66 ± 7 years) and control subjects (n = 31; age 66 ± 5 years) walked 30, 40, or 50 km/d for 4 consecutive days. Muscle injury markers (lactate dehydrogenase, creatine kinase, myoglobin, cardiac troponin I, and N-terminal pro-brain natriuretic peptide), muscle performance, and reported muscle symptoms were assessed at baseline and after exercise. Leukocyte CoQ10 was measured at baseline.

RESULTS All muscle injury markers were comparable at baseline ($P > 0.05$) and increased following exercise ($P < 0.001$), with no differences in the magnitude of exercise-induced elevations among groups ($P > 0.05$). Muscle pain scores were higher at baseline in symptomatic statin users ($P < 0.001$) and increased similarly in all groups following exercise ($P < 0.001$). Muscle relaxation time increased more in symptomatic statin users than in control subjects following exercise ($P = 0.035$). CoQ10 levels did not differ among symptomatic (2.3 nmol/U; IQR: 1.8-2.9 nmol/U), asymptomatic statin users (2.1 nmol/U; IQR: 1.8-2.5 nmol/U), and control subjects (2.1 nmol/U; IQR: 1.8-2.3 nmol/U; $P = 0.20$), and did not relate to muscle injury markers, fatigue resistance, or reported muscle symptoms.

CONCLUSIONS Statin use and the presence of statin-associated muscle symptoms does not exacerbate exercise-induced muscle injury after moderate exercise. Muscle injury markers were not related to leukocyte CoQ10 levels. (Exercise-induced Muscle Damage in Statin Users; NCT05011643) (J Am Coll Cardiol 2023;81:1353-1364)

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The authors attest they are in compliance with human studies committees and animal welfare regulations of the authors' institutions and Food and Drug Administration guidelines, including patient consent where appropriate. For more information, visit the Author Center.

Manuscript received November 29, 2022; revised manuscript received January 24, 2023, accepted January 30, 2023.

ISSN 0735-1097/$36.00 https://doi.org/10.1016/j.jacc.2023.01.043

ABBREVIATIONS AND ACRONYMS

CK = creatine kinase

CoQ10 = coenzyme Q10

CS = citrate synthase

cTnI = cardiac troponin I

LDH = lactate dehydrogenase

MVC = maximal voluntary contraction

mVO_2 = muscle oxygen consumption

NIRS = near-infrared spectroscopy

NT-proBNP = N-terminal probrain natriuretic peptide

SAMS = statin-associated muscle symptoms

Statins, physical activity, and their combination reduce initial and subsequent cardiovascular disease (CVD) events.[1,2] Statins are well-tolerated, but can cause statin-associated muscle symptoms (SAMS).[3] Physical activity may exacerbate SAMS, resulting in decreased physical activity or statin nonadherence.[4,5] Indeed, eccentric or vigorous exercise augments creatine kinase (CK) levels to a larger extent in statin users compared with nonstatin users,[6,7] suggesting that statins exacerbate exercise-induced skeletal muscle injury. It is unclear, however, if statins also increase muscle injury markers after moderate-intensity exercise, because moderate exercise does not exacerbate muscle symptoms and improves muscle performance in statin users.[8]

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The mechanisms underlying SAMS and statin-induced muscle injury are poorly understood. Statins inhibit 3-hydroxy-3-methyl-glutaryl-coenzyme A reductase, thereby decreasing mevalonate production and its downstream products including coenzyme Q10 (CoQ10), an essential cofactor of the mitochondrial electron transport chain. Mitochondrial dysfunction caused by reduced CoQ10 levels could predispose to muscle injury.[9] Exercise, especially high-intensity exercise, increases energy demands and could produce muscle fatigue and damage if energy production is compromised.[10] This mechanism is supported by the observation that mitochondrial dysfunction is more pronounced in patients with SAMS.[11] To our knowledge, the relationship between CoQ10 levels and exercise-induced muscle injury after prolonged moderate-intensity exercise has not been examined.

We sought to compare the impact of moderate-intensity exercise on muscle injury between symptomatic and asymptomatic statin users and nonstatin using control subjects. We also examined the association between CoQ10 levels in leukocytes and exercise-induced muscle injury and reported muscle symptoms. We hypothesized that statins would not exacerbate muscle injury after moderate-intensity exercise and that higher CoQ10 levels would be associated with less muscle injury and fewer reports of muscle symptoms.

METHODS

PARTICIPANTS. Statin users and nonstatin-using control subjects participating in the 102nd edition of the Nijmegen Four Days Marches were recruited via the Nijmegen Exercise Study database[12] and social media. Participants walk 30, 40, or 50 km/d at a self-selected pace for 4 consecutive days. Statin users were included if they had used statins continuously for ≥3 months before study participation. Statin dose potency was standardized to "atorvastatin" equivalence with 1 atorvastatin equivalent = 5 mg of atorvastatin = rosuvastatin 1.25 mg = simvastatin 10 mg = lovastatin 20 mg = pravastatin 20 mg.[13,14] Statin users were considered symptomatic or asymptomatic based on the presence, localization, and onset of muscle cramps, pain, and/or weakness, using the statin myalgia clinical index score.[15] Exclusion criteria included diabetes mellitus, hypothyroidism or hyperthyroidism, known hereditary skeletal muscle defects, other diseases known to cause muscle symptoms (eg, Parkinson or rheumatic diseases), and the use of CoQ10 supplementation. Participants provided written informed consent as approved by the medical ethics committee of the Radboud University Medical Center. This trial is registered within ClinicalTrials.gov (NCT05011643).

STUDY PROCEDURES. Baseline measurements were performed 1 or 2 days before the walking event. Post-exercise measurements were performed after the finish of the first, second, and third walking day. Weight, height, and waist circumference were measured at baseline. Weight was also measured before and after each walking day to assess hydration. Start and finish times were used to estimate exercise duration and walking speed. Heart rate was measured every 5 km during the first walking day using a 2-channel electrocardiographic chest band (Polar Electro Oy). Exercise intensity was calculated as average exercise heart rate divided by estimated maximum heart rate ($208 - 0.7 \times$ age).[16] The SQUASH (Short Questionnaire to Assess Health-enhancing physical activity) was used to determine physical activity at baseline.[17] Pain-rating index (PRI) and quality of life index (QoLI) scores were measured at baseline using the McGill Pain Questionnaire.[18] The more compact and applicable Brief Pain Inventory (BPI) and Brief Fatigue Inventory (BFI) were used to measure muscle pain and fatigue at baseline and postexercise to assess exercise-induced changes in reported muscle symptoms.[19,20] All questionnaires were filled in by the participants under supervision of a researcher.

LABORATORY ANALYSIS. Nonfasting venous blood was drawn from an antecubital vein at baseline to measure lipid profiles, vitamin D3, and muscle injury markers, including lactate dehydrogenase (LDH), CK, myoglobin, cardiac troponin I (cTnI) and N-terminal

pro-brain natriuretic peptide (NT-proBNP). Post-exercise blood samples were obtained within 1 hour of the finish on the walking days (postexercise). Serum was separated by centrifugation, aliquoted, and stored at −80 °C until analysis. Lipid profiles, CK, myoglobin, and LDH were measured by the Atellica CH Analyzer (Siemens Healthcare Diagnostics Inc). Serum 25-hydroxyvitamin D was measured by liquid chromatography coupled to tandem mass spectrometry detection (Waters Chromatography B.V.). cTnI was measured by a high-sensitivity assay (Attelica IM TnIH, Siemens Healthcare Diagnostics Inc).[21] NT-proBNP was measured by the Atellica IM NT-proBNP assay (Siemens Healthcare Diagnostics Inc). cTnI and NT-proBNP concentrations below the limit of quantification were given the limit of quantification value for cTnI (2.5 ng/L) and NT-proBNP (4 pmol/L).

LEUKOCYTE ISOLATION AND MITOCHONDRIAL FRACTION PREPARATION. Leukocytes were immediately isolated from the heparinized blood sample by the modified method of Skoog and Beck[22]: blood was mixed with 3% (w/v) dextran in 0.9% (w/v) NaCl in a ratio of 5:1 and incubated for 45 minutes at room temperature. The leukocyte-rich supernatant was removed and centrifuged at 694 *g* for 8 minutes. Contaminating erythrocytes were lysed by adding Milli-Q to the cell fraction. After 90 seconds, 2.7% (w/v) NaCl (2:1 [v/v] ratio) was added to regain an isotonic solution. After centrifugation for 8 minutes at 694 *g*, cells were washed 3 times with 0.9% (w/v) NaCl and stored at −80 °C. To obtain mitochondrial fractions, cell pallets were resuspended in phosphate-buffered saline and sonicated for 10 seconds on ice. Leukocytes were then resuspended in a buffer containing 730 mmol/L Tris and 416 mmol/L sucrose (pH 7.4) and centrifuged at 600 *g* for 10 minutes. The supernatant was removed and centrifuged at 14,000 *g* for 20 minutes, and the mitochondrial enriched fraction was resuspended in 0.1 mol/L Tris (pH 7.4).

CoQ10 ANALYSIS. CoQ10 levels (ubiquinone) were determined in leukocyte mitochondrial fractions, with coenzyme Q9 as the internal standard. [23] Samples were dissolved in ethanol (1:4 [v/v] ratio); the sample was dissolved in hexane (1:4 [v/v] ratio) and centrifuged at 2,300 *g* for 5 minutes. The upper layer was removed and evaporated to dryness under a nitrogen stream and the residue was dissolved in ethanol. CoQ10 was measured using a Nexera ultra-high-performance liquid chromatography (Shimadzu). CoQ10 levels were determined using a calibration curve in each experimental run. Subsequently, CoQ10 levels were normalized to citrate synthase (CS), a marker of mitochondrial mass. CS activity was measured in the mitochondrial enriched fractions.[24]

TABLE 1 Subject Characteristics

	Symptomatic (n = 35)	Asymptomatic (n = 34)	Control (n = 31)	*P* Value
Male/female	28/7	28/6	21/10	0.33
Age, y	64 (59-67)	68 (64-71)	68 (63-70)	0.010[a]
Statin type				0.19
Simvastatin	13 (37.1)	21 (61.8)	NA	
Atorvastatin	12 (34.3)	6 (17.6)	NA	
Rosuvastatin	7 (20.0)	4 (11.8)	NA	
Pravastatin	3 (8.6)	2 (5.9)	NA	
Fluvastatin	0 (0.0)	1 (2.9)	NA	
Atorvastatin equivalents[b]	4 (2-8)	4 (2-4)	NA	0.16
Duration statin therapy, mo	60 (36-120)	96 (57-132)	NA	0.19
Body mass index, kg/m^2	26.7 ± 3.2	26.4 ± 3.3	26.4 ± 3.7	0.92
Waist circumference, cm	96.1 ± 11.3	97.1 ± 11.2	95.7 ± 10.5	0.88
Subcutaneous adipose tissue,[c] mm	6.7 ± 1.6	6.2 ± 2.2	7.5 ± 2.1	0.28
Daily walking distance, km				0.13
30	16 (45.7)	22 (64.7)	21 (67.7)	
40	15 (42.9)	12 (35.3)	8 (25.8)	
50	4 (11.4)	0 (0.0)	2 (6.5)	
Walking duration,[d] h	8.1 ± 1.7	8.0 ± 0.9	7.6 ± 1.4	0.52
Walking speed,[d] km/h	4.6 ± 0.6	4.2 ± 0.6	4.5 ± 0.9	0.07
Exercise intensity, %	64.2 ± 9.3	63.9 ± 8.6	67.6 ± 7.2	0.17
Change in body mass,[d] %	−1.4 ± 0.6	−1.5 ± 0.6	−1.4 ± 1.1	0.91
Pain rating index, AU	20.4 (0-31.6)	0 (0-19.4)	0 (0-23.4)	0.018 [a,e]
Quality-of-life index, AU	1.5 (0.0-3.3)	0.0 (0.0-2.0)	0.0 (0.0-1.0)	0.050
Total physical activity, MET min/wk	4,893 (2,635-6,990)	4,629 (2,796-8,488)	4,272 (3,039-6,828)	0.70
Total cholesterol, mmol/L	4.0 ± 0.6	4.4 ± 0.7	5.7 ± 1.2	<0.001[e,f]
LDL cholesterol, mmol/L	2.0 ± 0.5	2.1 ± 0.6	3.3 ± 0.9	<0.001[e,f]
HDL cholesterol, mmol/L	1.3 ± 0.3	1.4 ± 0.3	1.5 ± 0.4	0.016[e]
Vitamin D3, nmol/L	108.8 ± 31.0	112.1 ± 24.7	111.7 ± 20.7	0.85

Values are n, median (IQR), n (%), or mean ± SD. Symptomatic indicates symptomatic statin users; asymptomatic, asymptomatic statin users. When significant differences between groups were found, pairwise comparisons were made by Bonferroni post-hoc testing. [a]Prevalues significantly different between symptomatic and asymptomatic statin users ($P < 0.05$). [b]One atorvastatin equivalent (5 mg) = rosuvastatin 1.25 mg = simvastatin 10 mg = lovastatin 20 mg = pravastatin 20 mg. [c]Subcutaneous adipose tissue thickness at the site of application of the NIRS sensor was assessed using ultrasound. [d]Values represent measurements on first walking day. [e]Prevalues significantly different between symptomatic statin users and control subjects ($P < 0.05$). [f]Prevalues significantly different between asymptomatic statin users and control subjects ($P < 0.05$).

AU = arbitrary units; HDL = high-density lipoprotein; LDL = low-density lipoprotein; MET = metabolic equivalent of task; NA = not applicable.

MUSCLE FUNCTION MEASUREMENTS. Handgrip strength of the dominant hand was measured at baseline and postexercise.[25] Additional muscle function measurements were performed in a subgroup of 16 symptomatic and 16 asymptomatic statin users and 18 control subjects 1 to 2 weeks before the walking event and within 1 hour postexercise. Near-infrared spectroscopy (NIRS) was used to measure the recovery of muscle oxygen consumption (mVO_2) after gastrocnemius muscle exercise, as a proxy of skeletal muscle mitochondrial capacity.[26] The 40-mm channel was used for analysis. The subcutaneous adipose

TABLE 2 Statin Treatment Used by Number of Participants

Type of Statin	Dose (mg/d)	Symptomatic (n = 35)	Asymptomatic (n = 34)
Simvastatin	10	1	1
	20	4	9
	40	8	11
Atorvastatin	10	4	0
	20	2	4
	40	6	1
	80	0	1
Rosuvastatin	5	1	3
	10	4	1
	20	2	0
Pravastatin	20	1	1
	40	2	1
Fluvastatin	80	0	1

tissue thickness at the site of application of the NIRS sensor was assessed using ultrasound (Table 1). Maximal voluntary contraction (MVC) and muscle fatigue resistance were determined in the quadriceps femoris of the dominant leg.[11] Muscle fatigue resistance was analyzed by calculating peak force and half relaxation time (RT) during repetitive electrical stimulation. Muscle fatigue resistance was also determined postexercise with the electrical current established during the baseline test.

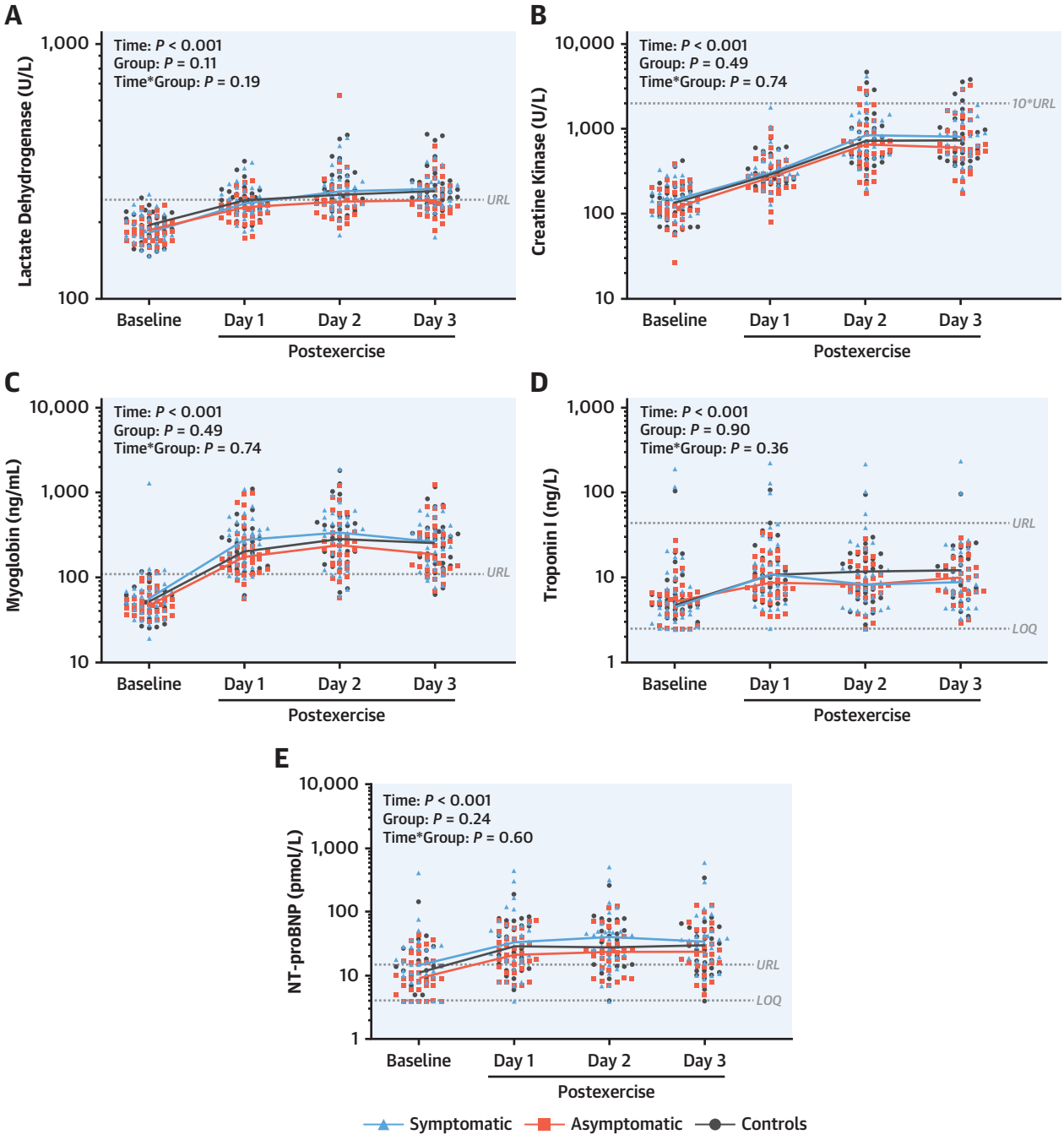
STATISTICAL ANALYSIS. Continuous variables were reported as mean ± SD or median (IQR). Categorical variables were reported in proportions and tested by the chi-square test. Differences at baseline were assessed by 1-way analysis of variance for normally distributed variables, and by Kruskal-Wallis 1-way analysis of variance for skewed variables. Changes within groups were assessed by paired Student's *t*-tests. Group comparisons of parameters measured at multiple time points were assessed by 2-way repeated measures analysis of variance. When data was not normally distributed, natural logarithmic transformation was applied. Bonferroni post hoc test were used to identify specific pairwise comparisons. Pearson or Spearman correlation analyses were conducted to identify correlations between variables for normally distributed and skewed variables, respectively. Statistical significance was set at $P < 0.05$. Analyses were performed using SPSS 28.0 (IBM).

RESULTS

PARTICIPANTS. In total, 35 symptomatic, 34 asymptomatic statin users, and 31 nonstatin-using control subjects participated in this study. Symptomatic statin users (age 64 years; IQR: 59-67 years; 80% men) were slightly younger compared with asymptomatic statin users (age 68 years; IQR: 64-71 years; 82% men), but age was not significantly different between symptomatic statin users and control participants (age 68 years; IQR: 63-70 years; 68% men) (Table 1). Statin type, "atorvastatin" equivalence, and duration of statin therapy were not different between the statin groups (Tables 1 and 2). There were no differences in body mass index, waist circumference, physical activity levels, or vitamin D3 among the groups at baseline (Table 1). Three symptomatic and 2 asymptomatic statin users dropped out on the second or third walking day caused by physical symptoms other than muscle symptoms. Exercise duration, walking speed, and change in body mass were not different among the groups on any walking day (all; $P > 0.05$) and are displayed for the first walking day (Table 1). Participants walked at 65% ± 9% of their predicted maximum heart rate, with no differences among groups (Table 1).

EXERCISE-INDUCED MUSCLE INJURY. LDH, CK, myoglobin, cTnI, and NT-proBNP levels were not different among groups at baseline (Supplemental Table 1), increased in all groups after exercise (all, $P_{time} < 0.001$), but were not different among groups (all, $P_{time \times group} > 0.05$) (Figure 1). No participants had CK elevations >10 times the upper reference limit (URL) ("marked CK elevation") at baseline or after the first walking day, but 3 symptomatic and 2 asymptomatic statin users as well as 4 control participants had marked CK elevations after the second walking day ($P = 0.62$). All of these participants again had marked CK elevations after the third day, except for 1 symptomatic statin user whose CK levels decreased to <10 x URL and 1 symptomatic statin user who could not be phlebotomized ($P = 0.29$). Cardiac TnI concentrations exceeded the 99th percentile in 2 symptomatic statin users and 1 control subject at baseline ($P = 0.52$). One additional symptomatic statin user demonstrated cTnI levels exceeding the 99th percentile after the first walking day ($P = 0.21$). Participants had similar cTnI responses after the second and third walking day (Supplemental Table 1). NT-proBNP concentrations exceeded the URL in 44% of symptomatic, 33% of asymptomatic statin users, and 32% of control subjects at baseline ($P = 0.59$). The prevalence of NT-proBNP concentrations exceeding the URL increased to 82% in symptomatic, 71% in asymptomatic statin users, and 71% in control subjects after the first walking day ($P = 0.46$). Comparable cTnI and NT-proBNP levels were observed after the second and third walking days, with no differences among groups (Supplemental Table 1).

FIGURE 1 **Exercise-Induced Muscle Injury Markers**



Lactate dehydrogenase **(A),** creatine kinase **(B),** myoglobin **(C),** cardiac troponin I **(D),** and N-terminal pro-brain natriuretic peptide (NT-proBNP) **(E)** were measured before (baseline) the walking event and after the finish of the first, second, and third walking day (postexercise). **Symbols** represent individual values, and **solid lines** represent group median. *P* values are derived from analysis on log-transformed data. LOQ = limit of quantification; URL = upper reference limit.

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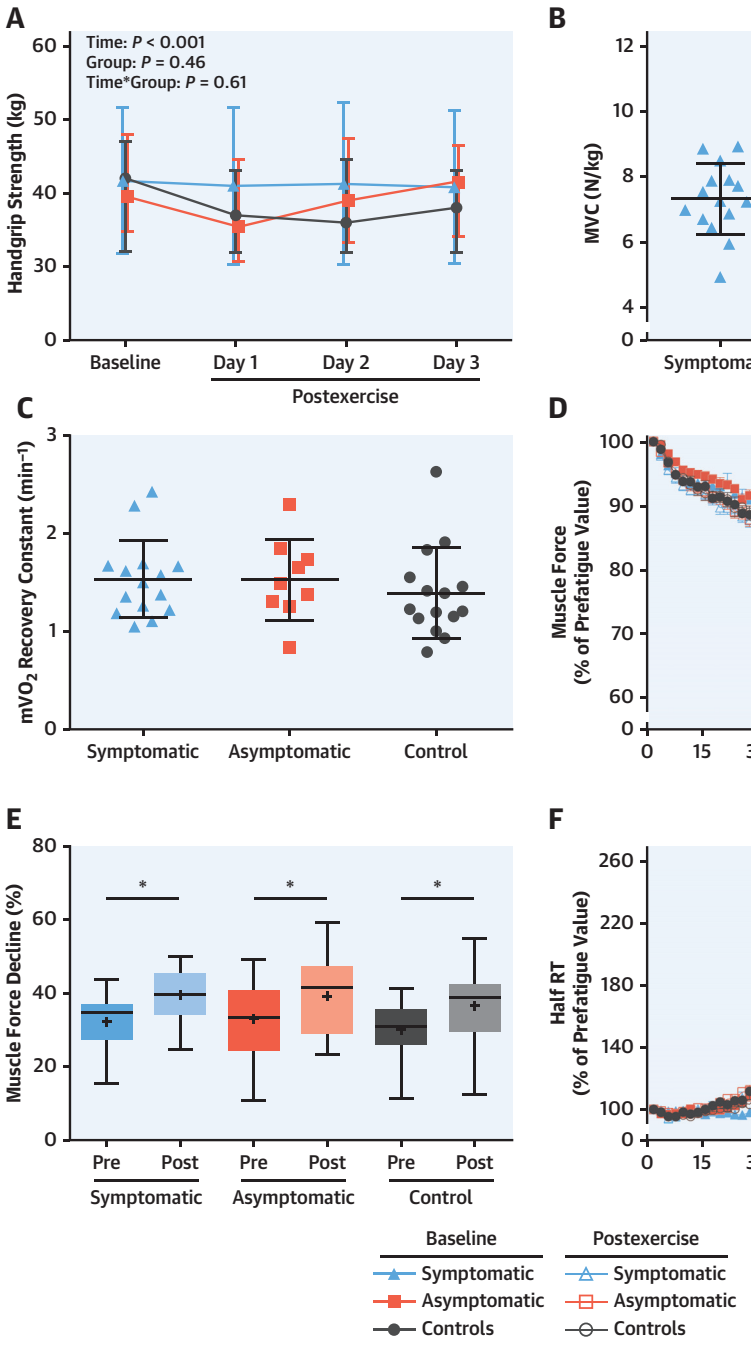
FIGURE 2 **The Effect of Moderate Exercise on Muscle Performance**



Handgrip strength **(A)** was measured at baseline and after finishing the walking days. MVC **(B)** and muscle oxygen consumption (mVO_2) **(C)** were measured at baseline. mVO_2 results of 1 symptomatic, 7 asymptomatic, and 3 control subjects were excluded. Values represent mean, whiskers represent SD. Muscle force **(D)** and half relaxation time (RT) **(F)** were determined at baseline (pre) and 1 day postexercise, and expressed as percentage of prefatigued value. Muscle force decline results were analyzed of 14 symptomatic, 13 asymptomatic, and 14 control subjects, and half RT results of 13 symptomatic, 12 asymptomatic, and 12 control subjects. Exclusions were because of poor data quality. Values represent mean, whiskers represent SEM. Muscle force decline **(E)** and increase in half RT **(G)** were calculated pre-exercise and postexercise. **Boxes** represent IQRs, **whiskers** 5th to 95th percentile, and the **horizontal line** and **cross** indicate median and mean. *Pre-measurement and postmeasurement significantly different. †Change between groups significantly different.

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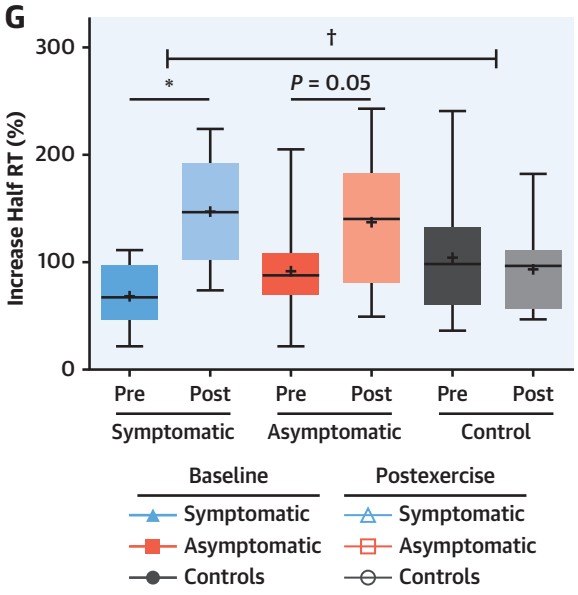
FIGURE 2 Continued



MUSCLE PERFORMANCE. There were no significant differences in handgrip strength, MVC, or mVO_2 among the groups at baseline (Figures 2A to 2C). Handgrip strength decreased in all groups post-exercise ($P_{time} < 0.001$), with no differences among groups ($P_{time \times group} = 0.61$) (Figure 2A). Muscle peak force (Figure 2D) and half RT (Figure 2F), determined during repetitive electrical stimulation and expressed as percentage of prefatigued value, were not different among groups at baseline ($P = 0.57$ and $P = 0.17$, respectively). The rate at which muscle peak force declined was significantly increased by exercise (7% ± 9%), with no differences among groups (Figure 2E). Half RT increased significantly in symptomatic statin users by 66% ± 39% ($P < 0.001$), tended to increase in asymptomatic statin users by 41% ± 65% ($P = 0.051$) and remained unchanged in control subjects following exercise ($P = 0.66$). The increase in half RT with exercise was significantly larger in symptomatic statin users compared with control subjects ($P = 0.035$) (Figure 2G).

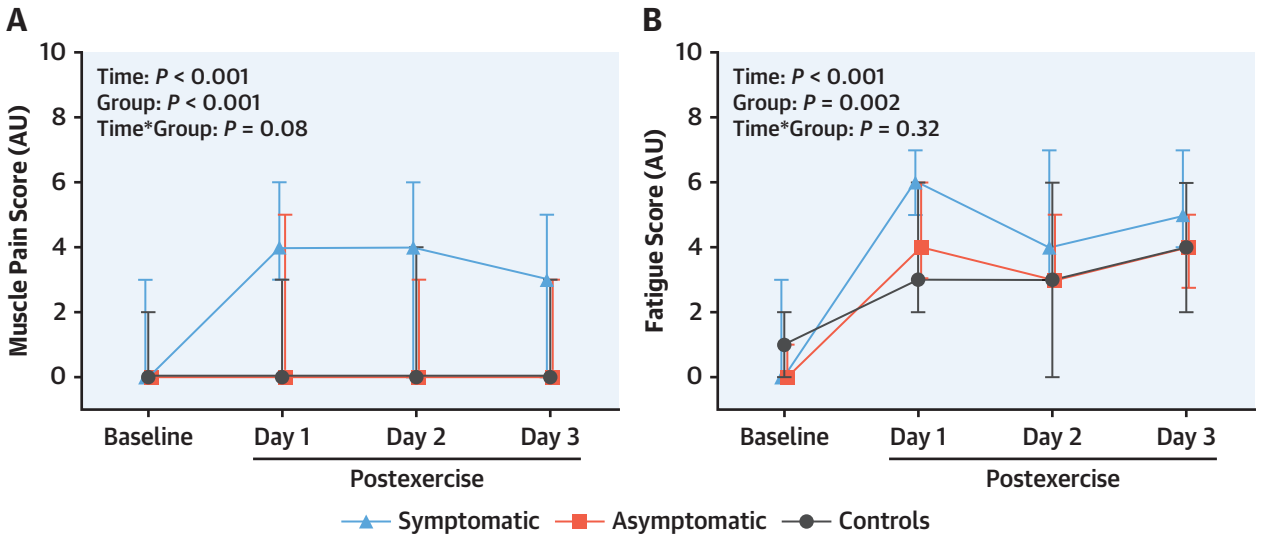
MUSCLE SYMPTOMS. Symptomatic statin users had higher PRI scores at baseline than asymptomatic statin users ($P = 0.046$) and control subjects ($P = 0.044$), and tended to have higher QoLI scores, implying lower quality of life ($P = 0.050$) (Table 1). Muscle pain scores were higher in symptomatic statin users compared with asymptomatic statin users and control subjects ($P_{group} < 0.001$), but increased similarly following exercise ($P_{time} < 0.001$) in all groups ($P_{time \times group} = 0.08$) (Figure 3A). Fatigue scores were higher in symptomatic statin users compared with asymptomatic statin users ($P_{group} = 0.004$) and control subjects ($P_{group} = 0.017$) at baseline, and increased similarly with exercise ($P_{time} < 0.001$) in all groups ($P_{time \times group} = 0.32$) (Figure 3B).

CHOLESTEROL LEVELS AND CoQ10. Total cholesterol levels were higher in control subjects compared with symptomatic and asymptomatic statin users ($P_{group} < 0.001$) and decreased with exercise ($P_{time} < 0.001$) in all groups ($P_{time \times group} = 0.33$) (Figure 4A). CoQ10 levels were not different between symptomatic (2.4 ± 0.7 nmol/U), asymptomatic statin users (2.3 ± 0.7 nmol/U), and control subjects (2.1 ± 0.5 nmol/U; $P = 0.20$) (Figure 4B) at baseline. CS activity did not differ among symptomatic (68.4 mU/mL; IQR: 47.4-99.0 mU/mL), asymptomatic statin users (65.5 mU/mL; IQR: 46.4-94.7 mU/mL), and control subjects (62.9 mU/mL; IQR: 48.6-91.5 mU/mL; $P = 0.54$) at baseline. There were no correlations between CoQ10 levels and muscle injury markers at baseline or after exercise (all; $P > 0.05$). There were also no correlations between CoQ10 levels and muscle fatigue resistance or muscle pain scores (all; $P > 0.05$).

DISCUSSION

Statin users may be more susceptible to exercise-induced muscle injury caused by mitochondrial dysfunction. Previous studies have shown that

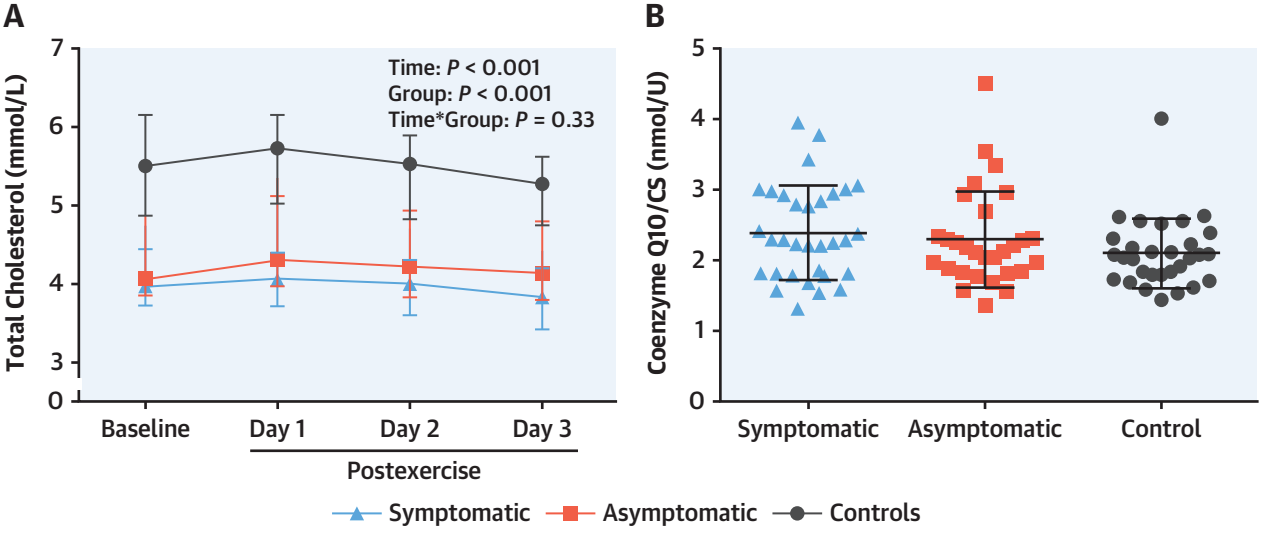
FIGURE 3 The Effect of Moderate Exercise on Muscle Symptoms



Muscle pain scores **(A)** and fatigue scores **(B)** were measured before (baseline) the walking event and after the finish of the first, second, and third walking day (postexercise). Values represent median and error bars represent interquartile (25th to 75th percentile) ranges.

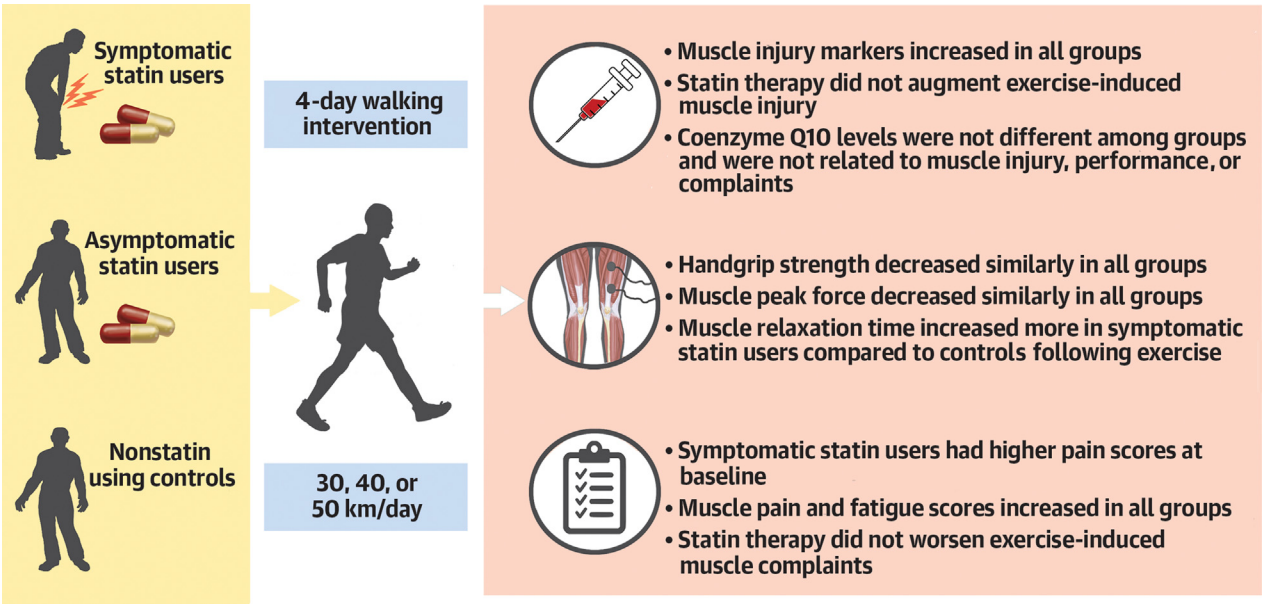
eccentric or vigorous exercise augments CK levels to a larger extent in statin users compared with statin nonusers. The present study demonstrated that statins did not exacerbate muscle injury markers, including LDH, CK, myoglobin, cTnI and NT-proBNP, after prolonged moderate-intensity exercise. Muscle injury markers were not related to leukocyte CoQ10 levels. Postexercise muscle strength decreased in all groups, but muscle relaxation time increased more in symptomatic statin users compared with control

FIGURE 4 Plasma Cholesterol Levels and Coenzyme Q10



(A) Total cholesterol levels were measured before (baseline) the walking event and after the finish of the first, second, and third walking day (postexercise). Values represent median and error bars represent IQR (25th to 75th percentile). *P* values are derived from analysis on log-transformed data. **(B)** Coenzyme Q10 was measured before (baseline) the walking event and was normalized to citrate synthase activity. Values represent mean and whiskers represent SD.

CENTRAL ILLUSTRATION **Exercise-Induced Muscle Injury in Chronic Statin Users**



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Statin use and the presence of statin-associated muscle symptoms does not augment exercise-induced muscle injury and reported muscle symptoms after prolonged moderate-intensity exercise. Muscle injury markers, performance, and reported muscle symptoms were not related to leukocyte coenzyme Q10 levels.

subjects. Even though muscle pain and fatigue scores were higher in symptomatic statin users at baseline, the increase in muscle symptoms after exercise was similar among the groups (Central Illustration). These results demonstrate that prolonged moderate-intensity exercise is safe for statin users and can be performed by statin users to maintain a physically active lifestyle and to derive its cardiovascular health benefits.

MUSCLE INJURY MARKERS. Prior research suggests that statins exacerbate muscle injury with eccentric and/or high-intensity exercise as evidenced by greater postexercise CK increases compared with control subjects.[6,7] Our results demonstrate that prolonged moderate-intensity exercise does not increase markers of muscle injury in statin users compared with nonstatin-using control subjects. Hence, the type, duration, and intensity of exercise may determine exercise-induced muscle injury, because these factors produce different mechanical and metabolic effects on skeletal muscle.[27,28] Statin users' skeletal muscle fibers are more susceptible to injury.[10] Therefore, moderate-intensity exercise may be more suitable for statin users and provoke less muscle injury than high-intensity or eccentric exercise.

We also measured cTnI and NT-proBNP levels to evaluate myocardial injury. cTnI and NT-proBNP release after exercise was not different among symptomatic and asymptomatic statin users and nonstatin-using control subjects. These results are consistent with studies that reported no increase in myocardial injury markers with statin therapy after marathon running or prolonged walking.[29,30] The contrast that statins could potentiate skeletal muscle but not cardiac muscle injury after eccentric or high-intensity exercise may be partly explained by different effects of statins on the skeletal muscle ryanodine receptor (RYR1) compared with their effect on the cardiac muscle RYR (RYR2).[31] RYR1 seems especially susceptible to the reactive oxygen species produced by statin therapy,[32] which could potentiate calcium release from the mitochondria and sarcoplasmic reticulum, ultimately leading to myocellular damage. Interestingly, moderate-intensity exercise reverses the sarcoplasmic reticulum calcium leak in rodent skeletal muscle, which suggests a protective effect of moderate-intensity exercise on statin-induced muscle injury.[31] Further research is

required to examine if moderate and vigorous exercise have different effects on RYR1 in human skeletal muscle.

Total cholesterol concentrations decrease within 2 hours after eccentric exercise and can persist for at least 48 hours,[33] possibly because cholesterol is used to repair cell membranes.[34] Total cholesterol could also be reduced because of increased uptake for an increased production in cortical hormones related with exercise.[35] Total cholesterol levels decreased similarly in all groups after exercise in the present study suggesting that cell damage was comparable among the groups. Moderate reductions in total cholesterol also occurred in the statin users, despite their lower baseline cholesterol values.

MUSCLE PERFORMANCE. There were no baseline differences in handgrip strength and MVC among the groups, and handgrip strength decreased in all groups following exercise. This agrees with most[36,37] but not all[38] studies reporting that statins do not alter muscle strength or performance. We have used repetitive involuntary contractions to produce muscle fatigue and have suggested that this may be a more sensitive technique to detect changes in muscle performance in statin users.[11] We did not find differences in muscle force decline or prolongation of relaxation time at baseline in the present study, but this may be because our participants were exercise trained.[8] However, the increase in muscle relaxation time was larger in symptomatic statin users compared with control subjects after exercise, with asymptomatic statin users showing intermediate values. The larger muscle fatigability in symptomatic statin users could be caused by disturbances in calcium homeostasis, energy metabolism, or muscle fiber distribution.[8,11,39] We could not obtain muscle biopsy specimens during the walking event to elaborate on these underlying mechanisms, but we did use NIRS to characterize skeletal muscle mitochondrial capacity and did not find differences in mVO_2 among the groups. We are unaware of previous studies that have measured the recovery of muscle oxygen consumption after exercise with NIRS in statin users. NIRS measurements have been able to detect differences in mitochondrial muscle oxygen consumption between high-fitness and low-fitness participants[40] and between young (age 19-25 years) and older (age 65-71 years) healthy men.[41] It is possible that NIRS is not sensitive enough to detect small changes in mitochondrial function associated with statin therapy. Muscle pain and fatigue scores increased in all groups after exercise with no differences among groups, which is consistent with our recent observation that a moderate-intensity exercise program is feasible for statin users.[8]

COENZYME Q10. CoQ10 depletion has been suggested to impair energy metabolism in statin users and to contribute to SAMS. Lower CoQ10 levels with statin therapy have been found primarily in blood plasma levels.[42] CoQ10 is transported in plasma in lower-density lipoproteins making it possible that lower CoQ10 blood levels simply reflect lower LDL levels from statin therapy.[43] We measured CoQ10 levels in mitochondria from leukocytes, because mononuclear cell and skeletal muscle cell CoQ10 levels are correlated ($r = 0.89$; $P < 0.02$).[44] We did not find differences in CoQ10 levels among the groups, and CoQ10 status was not related to muscle performance, reported muscle symptoms, or markers of injury. However, leukocytes are metabolically heterogenous and smaller differences in CoQ10 levels may not be detected in leukocytes as a result of both methodological and biological variations. Directly measuring CoQ10 levels in muscle samples is preferred but biopsies were not obtained in our study. Previous studies of intramuscular CoQ10 levels during statin therapy have shown reductions,[45] increases,[46] or no change,[47] but were only performed in asymptomatic[45,46] or symptomatic statin users,[47] included only statin-naïve participants,[45,46] and had small sample sizes of only 18 to 44 participants.[45-47]

STUDY LIMITATIONS. The present study shows that prolonged moderate-intensity exercise is safe for statin users, but the findings of this study cannot be directly extrapolated to other exercise modalities. We studied individuals who were capable of walking 30, 40, or 50 km daily for 4 consecutive days, and our participants with SAMS remained on their statin despite their reported muscle symptoms. Different results could occur with participants who are less physically capable, in those who are less exercise trained, and in SAMS patients with more disabling symptoms. Our participants used various statins (**Table 2**), which could have affected SAMS[3] and our results, although differences in statin type and dose were not statistically significant and differences in statins, such as hydrophilicity, do not appear to be as important as initially thought.[48-50] We did not obtain muscle biopsy specimens so we could not directly assess skeletal muscle mitochondrial function and muscular CoQ10 levels, although NIRS and leukocyte CoQ10 levels are accepted surrogates for muscle mitochondrial function and intramuscular CoQ10 levels, respectively.

CONCLUSIONS

Statin therapy does not augment exercise-induced muscle injury associated with prolonged moderate-intensity exercise. Muscle pain and fatigue scores increased similarly in all groups with exercise and were not worsened by statins. This study demonstrated that habitually active statin users can engage in prolonged moderate-intensity exercise without exacerbating skeletal muscle injury and reinforces the recommendation to combine statin therapy with a physically active lifestyle. This is an important strategy in the prevention of cardiovascular diseases.

ACKNOWLEDGMENTS The authors thank Bregina Kersten and Karina Horsting-Wethly for their technical assistance, as well as the participants for their enthusiasm and dedication in this study.

FUNDING SUPPORT AND AUTHOR DISCLOSURES

Dr Allard is financially supported by a grant from the Radboud Institute for Health Sciences. All other authors have reported that they have no relationships relevant to the contents of this paper to disclose.

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PERSPECTIVES

COMPETENCY IN PATIENT CARE AND PROCEDURAL SKILLS: Statin therapy does not augment exercise-induced muscle injury or reported muscle symptoms, and moderate-intensity exercise is safe for statin users with or without SAMS.

TRANSLATIONAL OUTLOOK: Further studies are needed to assess the relationship between intramuscular concentration of CoQ10 and muscle performance in patients taking statin medications.

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KEY WORDS coenzyme Q10, moderate-intensity exercise, muscle injury, muscle performance, statin-associated muscle symptoms

APPENDIX For a supplemental table, please see the online version of this paper.