

Exercise Medicine

ONLINE MANUSCRIPT SUBMISSION

Title: Statins do not impair fat metabolism during moderate-intensity exercise in dyslipidemic adults

Type of Manuscript: Research Article

Running Title: Statins, fat metabolism and exercise

Abstract

Objective: Some lipid-lowering agents, for example, nicotinic acid and fibrates, decrease an individual's ability to metabolise fat during exercise. However, it is unclear whether statins affect fat metabolism during exercise in patients. This study investigated whether fatty acid oxidation is impaired in a dyslipidemic population, while walking at a moderate intensity. Methods: Patients (n=16), walked for 45 minutes on a treadmill at 50% of their estimated $\dot{V}O_{2max}$, in the absence and presence of their prescribed statin. Fat metabolism was investigated by examining respiratory data, and circulating plasma glycerol and free fatty acids. Results: Analysis of respiratory data indicated a progressive increase in fat oxidation over time, along with a decrease in carbohydrate oxidation, for all patients during exercise, in both the absence and presence of a statin ($P \leq 0.05$). The increase in the percent of energy derived from fat was further supported by the observation of a significantly progressive increase in circulating glycerol and free fatty acids during the exercise period. However no significant difference in the extent of change was observed when comparing the respiratory and biochemical response to physical activity in the absence and presence of the prescribed statin. Conclusions: There is no evidence of a negative impact of statins on the ability to metabolise fat as a fuel for moderate-intensity aerobic exercise. Given the importance of physical activity, this result encourages patients to participate in regular exercise without concern of premature fatigue.

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Abstract

Objective: Some lipid-lowering agents, for example, nicotinic acid and fibrates, decrease an individual's ability to metabolise fat during exercise. However, it is unclear whether statins affect fat metabolism during exercise in patients. This study investigated whether fatty acid oxidation is impaired in a dyslipidemic population, while walking at a moderate intensity.

Methods: Patients (n=16), walked for 45 minutes on a treadmill at 50% of their estimated $\dot{V}O_{2max}$, in the absence and presence of their prescribed statin. Fat metabolism was investigated by examining respiratory data, and circulating plasma glycerol and free fatty acids.

Results: Analysis of respiratory data indicated a progressive increase in fat oxidation over time, along with a decrease in carbohydrate oxidation, for all patients during exercise, in both the absence and presence of a statin ($P \leq 0.05$). The increase in the percent of energy derived from fat was further supported by the observation of a significantly progressive increase in circulating glycerol and free fatty acids during the exercise period. However no significant difference in the extent of change was observed when comparing the respiratory and biochemical response to physical activity in the absence and presence of the prescribed statin.

Conclusions: There is no evidence of a negative impact of statins on the ability to metabolise fat as a fuel for moderate-intensity aerobic exercise. Given the importance of physical activity, this result encourages patients to participate in regular exercise without concern of premature fatigue.

Key Words: adipose tissue, aerobic exercise, cholesterol-lowering drugs, fat oxidation, physical activity

47 **Introduction**

48 Lipid-lowering drugs are amongst the most commonly subsidized prescribed drugs world-wide,
49 including Europe and the United States (USA). In 2016, atorvastatin and simvastatin were ranked in
50 positions 1 and 7 in the USA [1]. In the same year, in Australia, atorvastatin and rosuvastatin were in
51 positions 1 and 3, with the number of Australians per thousand taking the drug daily being 53 for
52 atorvastatin and 34 for rosuvastatin [2].

53 Physiologically, these drugs inhibit liver enzyme HMG-CoA reductase, blocking cholesterol
54 synthesis. They are recognized as effective in the secondary prevention of cardiovascular disease and
55 all-cause mortality [3]. However, it is now well established that a healthy diet and regular physical
56 activity are valuable contributors to maintaining healthy blood lipids, with exercise improving
57 cardiovascular fitness; reducing all-cause mortality and the risk of coronary heart and cardiovascular
58 disease, stroke, type 2 diabetes mellitus (T2DM), and obesity. A recent meta-analysis highlighted that
59 physical activity alone is potentially as effective as many drug interventions on mortality outcomes in
60 coronary heart disease, stroke, heart failure, and prediabetes [4]. Despite this, there exists a bias against
61 exercise intervention with a subsequent progression towards lowering the threshold for pharmaceutical
62 treatment, and an increased likelihood that individuals embarking on an exercise regime are already
63 taking a cholesterol-lowering agent as a secondary or even primary prevention against cardiovascular
64 risk. To maximize the benefit of pharmaceutical intervention and exercise, it is important therefore, to
65 understand the effect of these drugs on the body's response to exercise.

66 Lipid stores, critical for fueling prolonged exercise, exist as triglycerides in adipose tissue and
67 muscle, and also as circulating triglycerides. Hormone-sensitive lipase (HSL), an intracellular enzyme in
68 adipose tissue, and lipoprotein lipase (LPL), an enzyme which hydrolyzes the triglyceride core of
69 circulating chylomicrons, both liberate free fatty acids (FFA) for use by exercising muscle. Any drugs
70 acting at these sites, could potentially alter FFA availability, resulting in increasing dependence on

71 muscle glycogen stores and earlier fatigue [5]. Studies in healthy volunteers undertaking exercise have
72 found that the lipid-lowering drugs, acipimox, and fibrates such as bezafibrate and gemfibrozil, have a
73 negative impact on fat oxidation [5,6]. This increases demand for carbohydrate (CHO), and is thought to
74 contribute to a reduction in exercise tolerance. However, no negative impact of simvastatin, fluvastatin
75 or atorvastatin has been observed in healthy exercising participants [5,6,7].

76 In contrast, the impact of statins on fat metabolism in patient cohorts is less clear. The available
77 research in patients undergoing exercise has produced conflicting results, with studies confounded by
78 the co-administration of a kaleidoscope of other cholesterol-lowering agents, or with β -blockers, which
79 inhibit the release of FFA from adipose tissue and complicate the interpretation of the data [8,9]. One
80 study in patients has reported reduced fat oxidation with atorvastatin, during lower-intensity exercise
81 [10]. Patients have also self-reported increased fatigue with exertion after taking simvastatin for a period
82 of 6 months [11]. In contrast, other studies have reported no negative impact of statins on fat metabolism
83 [12,13].

84 Given the importance of exercise in these patients, and the limited and conflicting data on the
85 interaction between cholesterol-lowering agents and physical activity, the current study investigated in a
86 dyslipidemic population, whether statins impair fatty acid oxidation during exercise of moderate-
87 intensity. This study is unique as it appears to be the first longitudinal investigation of fat metabolism in
88 both the absence and presence of a statin during exercise, within the same group of patients.

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93 **Method**

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95 **Participants**

96 The study engaged 16 Caucasian adults taking a statin for hypercholesterolemia. The study was
97 approved by the Human Research Ethics Committees from The University of New South Wales (UNSW)
98 (HREC05310) and the Sydney Adventist Hospital (HREC12/06) and all procedures were in accordance
99 with institutional guidelines. Participants were recruited by newspaper advertisements, and required
100 written permission from their medical practitioner to participate. Individuals also provided written
101 informed consent, and were aware of their right to withdraw at any time. Participants were considered
102 able to engage in physical activity comprising of a brisk walk, by satisfactorily completing a physical
103 activity readiness questionnaire (PAR-Q), as well as an additional questionnaire on family history,
104 medical history, present symptoms, and current pattern of physical activity [14,15]. Patients with T2DM
105 were excluded as this condition may influence fat metabolism. In addition, recruited participants were
106 not taking other medications which could knowingly interfere with fat metabolism.

107

108 **Experimental design**

109 Each participant made a total of three visits to the laboratory. All participants ceased lipid-lowering
110 medication for a minimum of three weeks prior to Visit 1. The purpose of Visit 1 was to (a) prescreen the
111 participant, (b) perform a submaximal exercise test on a treadmill to predict the participant's maximum
112 oxygen uptake ($\dot{V}O_{2max}$), and (c) calculate the individual treadmill settings (speed and gradient) which
113 would enable the participant to walk at a power output of 50% $\dot{V}O_{2max}$ in Visits 2 and 3. The latter were
114 identical to each other with the notable exception that Visit 2 continued in the absence of the drug and
115 Visit 3 in the presence of the drug. Visit 3 occurred after the participant had resumed their lipid-lowering

116 medication for a minimum of 3 weeks. Statin administration was verified verbally and by assessment of
117 plasma lipids measured at the end of the drug versus no drug phases, on the mornings of Visits 2 and 3.

118

119 **Visit 1: Prescreening and submaximal exercise test**

120 Resting heart rate (HR), blood pressure, anthropometric assessment (height, weight, body mass index
121 (BMI), waist and hip circumference), and percent body fat by bioelectrical impedance (Tanita BWB-800,
122 Tanita Corporation, Japan) and by skinfolds analysis (biceps, tripceps, subscapular, suprailiac and
123 abdominal) was measured [16].

124 Participants performed a submaximal exercise test on a treadmill to predict $\dot{V}O_{2max}$ and to
125 calculate the individual treadmill setting for each participant to exercise at 50% of their maximal
126 capability in Visits 2 and 3. Ventilation ($\dot{V}E$, l.min⁻¹), oxygen uptake ($\dot{V}O_2$, l.min⁻¹), expired carbon
127 dioxide ($\dot{V}CO_2$, l.min⁻¹) and respiratory exchange ratio (RER) ($\dot{V}CO_2/\dot{V}O_2$) were measured using a
128 metabolic cart (ParvoMedics TrueMax 2400). HR was monitored via an electrocardiogram (ECG) and a
129 HR monitor. Participants chose a comfortable walking speed at 0% gradient. With the speed constant,
130 exercise intensity was increased a further two times, by increasing the gradient of the treadmill belt
131 every 3-5 min after a stable exercise HR was recorded. The predicted maximum oxygen consumption
132 was calculated by plotting exercise HR against oxygen consumption, with a line of best fit extrapolated
133 to the point of each participant's maximal predicted HR [15]. The relative perceived exertion (RPE) [15]
134 and finger prick blood lactate, confirmed participants exercised at a submaximal level.

135

136 **Visits 2 and 3: Exercise at 50% $\dot{V}O_{2max}$ in the absence and presence of a statin**

137 The exercise trials occurred at Visit 2 (-statin) and Visit 3 (+statin). Visit 3 occurred after participants
138 had resumed medication for a minimum of three weeks. The two visits were otherwise identical.

139 Participants arrived at the laboratory after an overnight fast (water permitted). A forearm vein was

140 cannulated using a 20 gauge indwelling line (BD Insyte Autoguard, Becton Dickinson, UK). As the act

141 of venepuncture can increase some hormone levels by more than 50% [17], a stabilization period of 30
142 min was allowed after the cannula was inserted into the forearm vein, and before collection of a fasting
143 blood sample (6ml) into vacutainer tubes (Becton Dickinson, UK) containing ethylenediaminetetra-
144 acetic acid (EDTA). Fasting blood lipids, lactate and glucose were measured immediately. Participants
145 then consumed a low fat 1MJ CHO meal, consisting of Special K cereal and low fat dairy or lactose free
146 milk, followed by rest in a semi-inclined position. Participants commenced exercise 90 min after their
147 meal. They walked on the treadmill at an intensity of 50% of their calculated $\dot{V}O_{2max}$ (determined in
148 Visit 1), corresponding to a 'brisk walk' for 45 minutes. The treadmill speed for each participant was
149 that of Visit 1, and the treadmill gradient was determined using the formula: $\dot{V}O_2 = (0.1 \times S) + (1.8 \times S \times$
150 $G) + 3.5$; where $\dot{V}O_2$ is gross oxygen consumption $mL.kg^{-1}.min^{-1}$ at 50% of estimated $\dot{V}O_{2max}$, speed (S)
151 is in $metres.min^{-1}$ and G is percent grade expressed as a fraction [15]. The predetermined treadmill
152 settings (speed and gradient) for each individual participant remained constant throughout the exercise
153 session. VE , $\dot{V}O_2$, $\dot{V}CO_2$, and RER were measured to determine the proportion of CHO and fat
154 metabolism during exercise. Calculation of carbohydrate and fat oxidation was by the classical
155 stoichiometric equations of indirect calorimetry [18]; with carbohydrate oxidation (mg/min)
156 $= 4.58\dot{V}CO_2 - 3.2\dot{V}O_2$; and fat oxidation (mg/min) $= 1.7\dot{V}O_2 - 1.7\dot{V}CO_2$. HR and RPE were monitored.
157 Additional blood (each 6ml) was collected into EDTA-containing tubes immediately before exercise,
158 and at time 15, 30 and 45 minutes of exercise, for the immediate measurement of glucose and lactate.
159 Remaining blood samples were centrifuged (4 °C, 1560g, 10min, Heraeus Megafuge 1.0R, Hanau,
160 Germany) and plasma was stored at -86 °C prior to the analysis of glycerol and FFA.

161

162 **Biochemistry**

163 Lipids (total cholesterol (TC), triglycerides (TG), low density lipoprotein-cholesterol (LDL-C) and high
164 density lipoprotein-cholesterol (HDL-C)) (Cholestech LDX, CA, USA), glucose (HemoCue Glucose

201, Sweden), and lactate (Lactate Pro, ARKRAY, Inc., Japan) were measured in whole blood by reflectance photometry. Glycerol was measured in plasma using two colorimetric assays for comparison (Sigma, USA; and Cayman Chemical, USA) and the concentration determined at 550nm using an Expert Plus Microplate reader (Asys Hitech, Austria) and a Versa Max Microplate reader (Molecular Devices, USA). FFA were measured in plasma by colorimetric assay (Cell Biolabs, USA) at 570nm using the Versa Max Microplate reader.

171

172 **Food diary**

173 Participants completed a 3-day food diary, to calculate total caloric intake, and percentages of
174 macronutrient and micronutrient intake during two non-consecutive weekdays and one weekend day.
175 Data were entered into a nutritional database (Serve Nutrition Systems, Australia).

176

177 **Statistical Analysis**

178 Power calculations [19], based on a previous study [5] where participants walked at 50% $\dot{V}O_{2max}$ in the
179 presence or absence of a lipid-lowering agent, acipimox, determined that 11 participants would be
180 sufficient to detect a significant decrease in plasma glycerol, assuming a 0.05 significance level and 80%
181 power. The current study recruited 16 participants. All data are expressed as mean $\pm$ standard error of the
182 mean (SEM). The Student's paired *t*-test compared fasting blood lipids $\pm$ statin. A two-way repeated
183 measures ANOVA examined differences between the two within-subject factors (treatment and time) for
184 the physiological variables (HR, RPE, fat and CHO oxidation, % energy derived from fat and CHO,
185 RER, $\dot{V}O_2$, glucose, lactate, glycerol, and FFA), measured during exercise. Where a significant two-way
186 interaction was found (FFA), simple main effects examining differences between trials at each level of
187 time and treatment were determined using the General Linear Model (GLM) Repeated Measures
188 procedure. For cases without a significant two-way interaction, the main effect for treatment and time

189 were determined. Variables which did not meet Mauchly's test of sphericity were interpreted using a
190 Greenhouse-Geisser correction. For all data comparisons, unless indicated, significance is at $P \leq 0.05$. In
191 using the GLM Repeated measures procedure for the FFA data, a Bonferroni adjustment was applied for
192 the testing of multiple main effects, with a significance of $p \leq .05/3 = p \leq .016$. Analysis was conducted
193 using SPSS (SPSS Inc., USA).

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196 **Results**

197 **Patient characteristics**

198 The study engaged 13 males and 3 females, aged 39-82 years, taking a statin for hypercholesterolemia.
199 In addition to hyperlipidemia, some patients had cardiovascular conditions (hypertension, coronary
200 artery disease, heart murmur, aortic valve replacement, mild myocardial regurgitation, varicose veins),
201 and arthritis. Two participants, with the permission of their medical practitioner, ceased taking
202 metoprolol for the entire duration of the study as it interferes with fat metabolism [8].

203 Patients were non-smokers or previous smokers who had ceased smoking 15-35 years earlier.
204 Body composition data revealed that participants were on average overweight, had an increased disease
205 risk relative to weight and waist circumference, and a moderate disease risk based on waist-hip ratio [15].

206 Regarding physical activity, participants reported an average occupational activity intensity of
207 1.5 ± 0.2 (1=sedentary; 5=heavy labour), sport and leisure physical activity intensity of 6 ± 0.4 (1=very
208 light, 10=intense breathless), and exercise duration of 8 ± 1 hours per week. Activities included cycling,
209 swimming, board paddling, running, walking, dance, stair climbing, gym (circuit, weight training,
210 aerobic classes), tennis, rollerblading, and martial arts. Aerobic fitness, indicated by a predicted $\dot{V}O_{2\max}$,
211 was categorized as 'average' for males ($35 \pm 3 \text{ ml.kg}^{-1}.\text{min}^{-1}$) and 'fair' for females ($23 \pm 0.5 \text{ ml.kg}^{-1}.\text{min}^{-1}$)
212 [15]. The average percent daily consumption of protein and CHO was slightly higher, and lower, than
213 the recommended 10-15% and 50-60%, respectively. The average percent daily consumption of <30%
214 total fat was as recommended [20]. (Table 2)

215

216

217 **Lipid-lowering agents and blood lipoproteins**

218 Of the 16 patients, 11 were taking atorvastatin, with a daily dose range of 10-80mg (1=10mg, 6=20mg,
219 3=40mg, 1=80mg). Rosuvastatin, at 10 mg/day was prescribed for 1 patient, and the remaining 4

220 patients were administering simvastatin at a dose range of 10-40mg/day (1=10mg, 1= 20mg, 2=40mg).
221 Patients had been taking their drug for 3.5 ± 0.6 years (range 0-10 years). There was one newly diagnosed
222 individual who had not yet commenced his statin medication. All patients ceased taking their statin for
223 3.4 ± 0.2 weeks prior to their blood lipids analysis in Visit 2. Following this visit, patients recommenced
224 taking their medication for 26 ± 0.7 days. A significant decrease was observed in TC, LDL-C and
225 TC/HDL-C when taking a statin, compared with in the absence of the drug (Table 3). Though not
226 significant, a decrease in TG was also observed ($p=.073$).

227

228 **Energy expenditure and metabolism during exercise**

229 All participants completed the exercise tests. Exercise performed at 50% of $\dot{V}O_{2\max}$ for 45 minutes
230 required a total energy expenditure of 268 ± 20 kcal or 282 ± 21 kcal in the absence or presence of a statin,
231 respectively.

232 No two-way interaction (between treatment and time) was found for fat or CHO oxidation, %
233 energy derived from fat or CHO, RER, $\dot{V}O_2$, glycerol, glucose, lactate, HR, and RPE (Table 4). There
234 was a two-way interaction between treatment and time for FFA ($F(1.171, 12.879)=5.366$, $p=.033$,
235 $\epsilon=.58$) at 45 minutes ($p=.024$). However, this effect was disregarded following correction for the
236 testing of multiple main effects, with a Bonferroni adjustment, ($p<.016$), resulting in non-significance.

237 As there was no 2x3 interaction, the main effect for treatment (irrespective of time point) and the
238 main effect for time (irrespective of treatment) were examined. Regarding treatment, a difference
239 between the conditions ($\pm$ statin) was found for HR and $\dot{V}O_2$ ($p \leq 0.05$), with these variables lower in the
240 presence of the statin. The main effect of time was significant for all variables ($p \leq 0.05$).

241

242 **Discussion**

243 The current study has shown no evidence of an adverse effect of statins in dyslipidemic patients on the
244 ability to use fat as a metabolic fuel during a moderate-intensity 45-minute walk. The results in this
245 patient group are in agreement with previous research in healthy volunteers [5], where simvastatin
246 treatment had no impact on fat oxidation or circulating plasma free fatty acids, glycerol and glucose
247 during walking. This result is encouraging for patients, given that the importance of regular physical
248 activity is recognized as a positive contributor to improving and maintaining health [4]. Indeed, this
249 study supports positive messaging to patients taking statins; that they may engage in regular exercise.
250 This does to some extent counteract the recently suggested association of statins with an increased risk
251 of T2DM [21]. Furthermore, implementation of a healthy diet and regular aerobic exercise, while on
252 statin medication, may for some patients, enhance the possibility of a medically managed reduction in
253 their statin dose or even complete cessation of this medication. The current study provides reassurance
254 for both patients and their advising physicians, that it is possible to engage effectively in moderate-
255 intensity exercise without the prescribed statin compromising the ability to use fat stores as a fuel to
256 carry out the exercise session.

257 The lack of an adverse effect on fatty acid mobilization by statins may be due to the fact that
258 their action is thought to be largely confined to the liver [22], rather than adipose tissue. Preliminary
259 research by our group on ezetimibe (Ezetrol), a non-statin whose action is confined to inhibiting
260 cholesterol absorption from the intestine [23], has also shown no effect of this drug on fat metabolism
261 during 45 minutes of moderate-intensity walking (data not published). In contrast, acipimox and fibrates,
262 significantly inhibit HSL in adipose tissue, which results in inhibiting the release of FFA during exercise
263 from this site [5,6]. Fibrates may also reduce availability of FFA to skeletal muscle, by increasing the
264 uptake of FFA into liver with the induction of a fatty acid transporter protein [24].

265 The findings from this study, therefore make a valuable contribution to the somewhat confusing
266 information on statins and metabolism. It has been suggested that statins, by lowering cholesterol, may

267 affect mouse membrane calveolae, important in fatty acid transport [25]. Statins have been implicated in
268 impairing mitochondrial function, evidenced by increased lactate production, elevated intramuscular
269 lipid stores, decreased mitochondrial activity (measured by a decrease in cytochrome C activity) and
270 lower CoQ10 levels [26,27]. However, despite the above, it has been found that a decrease in circulating
271 CoQ10 does not result in a reduction in muscle CoQ10 [28]. Furthermore, reviews to date, have not
272 concluded that CoQ10 is required when taking statins [29,30].

273 Other conflicting results include reports in humans of increased resting RER with statins,
274 indicating a possible shift from fat to CHO metabolism [10,31]. A 12-week aerobic training study
275 measured a greater increase in $\dot{V}O_{2max}$ in a -statin, compared with +statin group. The authors reported a
276 decline in muscle citrate enzyme activity, suggesting that statins may affect mitochondria [32]. However,
277 in contrast to the above, there are reports of simvastatin therapy in patients having no effect on RER or
278 aerobic fitness ($\dot{V}O_{2max}$) suggesting no effect on fat metabolism and aerobic capacity [30]. Additionally,
279 a 30% increase in $\dot{V}O_{2max}$, indicating an increase in aerobic fitness, has been reported in patients on
280 rosuvastatin undergoing exercise training for 10 weeks [13].

281 The conflicting results in the literature, are due to many variables, including that of studying
282 different patient groups in either the presence or absence of the drug. The current study eliminated this
283 variable, adopting a longitudinal design, with all participants required to undertake exercise with and
284 without a statin. Based on previous research in healthy volunteers [5], the current study adopted a 3-
285 week treatment period verified by assessment of plasma lipids at the end of the drug versus no drug
286 phases. The treatment period significantly lowered TC and LDL-C, confirming that it was of sufficient
287 length to obtain the desired effect of lowering circulating lipids, enabling patients to serve as suitable
288 controls.

289 To minimize disruption to patients, the current study did not adopt a cross-over design, with all
290 patients experiencing the prescreening session and the first 45-minute walk in the absence of the drug,

and the second 45-minute walk in the presence of the drug. Additionally, researchers did not interfere with each individual patient's statin type and therapeutic dosage, to keep with their real-life situation, controlled by their physician. This, along with the age range of the patients, was not considered a limitation, as the strength of this study is in having each patient serve as their own effective control.

This study also concentrated on the effect of statins without the confounding variable of decreased blood glucose during exercise. The mean energy expenditure during exercise was 1.1MJ (282 kcal), well below the CHO energy reserve of 8-11MJ for a well-nourished adult [5,33]. Additionally, participants consumed a low fat 1MJ CHO meal 90 minutes prior to exercise. Blood glucose levels remained stable during both trials. The decreasing contribution of CHO and increasing contribution of fat as a fuel during the walk was similar in the $\pm$ statin exercise sessions. At 45 minutes of walking, fat oxidation peaked at an average of 0.28g/min.

The chosen modality and intensity of exercise, with a duration of 45 minutes, was adopted as it has been reported as manageable for patients and ideal for maximizing cardiovascular health benefits, significantly reducing morbidity and mortality [5,34]. Though it appears that statins do not impact fat metabolism when exercising under these conditions, future studies examining different exercise modalities, intensities and duration, are recommended.

There are many people world-wide on statins. Amongst this group are individuals who are considered capable of adopting a healthier lifestyle incorporating regular exercise. This study demonstrates that statins do not compromise fat metabolism during moderate-intensity aerobic exercise, and therefore do not contribute to premature fatigue and an inability of the patient to complete the exercise. More emphasis may therefore be placed on encouraging regular physical activity in patients to maximize the benefits of concomitant therapies such as pharmaceutical intervention.

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317

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321 University of New South Wales.

322

323 **Conflict of Interest**

324 The authors declare no conflict of interest exists with respect to research, authorship and publication.

325

326 **Author contribution**

327 MM and RG contributed to the conception of the study. MM designed the study, acquired, analysed and
328 interpreted the data, and drafted the manuscript. MM and RG critically revised the manuscript. Both
329 gave final approval and agree to be accountable for all aspects of work ensuring integrity and accuracy.

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References

1. LowestMed 2016. Top 50 Prescription drugs filled in the US in Q1 of 2016. [Updated 16 June, 2016]. <https://lowestmed.com/top-50-prescription-drugs-filled>. (Accessed 15 February, 2017)
2. NPS MedicineWise (2016) Top 10 Drugs 2015-16. *Aust Prescr* 2016;39:2205.
3. Naci H, Brugs JJ, Fleurence R, Tsoi B, Toor H, Ades AE. Comparative benefits of statins in the primary and secondary prevention of major coronary events and all-cause mortality: a network meta-analysis of placebo-controlled and active-comparator trials. *Eur J Prev Cardiol* 2013;20:641-57.
4. Naci H, Ioannidis JPA. Comparative effectiveness of exercise and drug interventions on mortality outcomes: metaepidemiological study. *BMJ* 2013;347:f5577.
5. Head A, Jakeman PM, Kendall MJ, Cramb R, Maxwell S. The impact of a short course of three lipid lowering drugs on fat oxidation during exercise in healthy volunteers. *Postgrad Med J* 1993;69:197-203.
6. Eagles CJ, Kendall MJ, Maxwell S. A comparison of the effects of fluvastatin and bezafibrate on exercise metabolism: a placebo-controlled study in healthy normolipidaemic subjects. *Br J Clin Pharmacol* 1996;41:381-87.
7. Chung J, Brass EP, Ulrich RG, Hiatt WR. Effect of Atorvastatin on energy expenditure and skeletal muscle oxidative metabolism at rest and during exercise. *Clin Pharmacol Ther* 2008;83:243-50.
8. Eagles CJ, Kendall MJ. The effects of combined treatment with β_1 -selective receptor antagonists and lipid-lowering drugs on fat metabolism and measures of fatigue during moderate intensity exercise: a placebo-controlled study in healthy subjects. *Br J Clin Pharmacol* 1997;43:291-300.
9. Fisher NM, Meksawan K, Limprasertkul A, Isackson PJ, Pendergast DR, Vladutiu GD. Statin therapy depresses total body fat oxidation in the absence of genetic limitations to fat oxidation. *J Inherit Metab Dis* 2007;30:388-99.
10. Limprasertkul A, Fisher NM, Awad AB, Pendergast DR. Statin therapy depresses fat metabolism in older individuals. *J Am Coll Nutr* 2012;31:32-8.
11. Golomb BA, Evans MA, Dimsdale JE, White HL. Effects of statins on energy and fatigue with exertion: results from a randomized controlled trial. *Arch Intern Med* 2012;172:1180-82.
12. Parker BA, Thompson PD. Effect of statins on skeletal muscle: Exercise, myopathy, and muscle outcomes. *Exerc Sport Sci Rev* 2012;40:188-94.
13. Coen PM, Flynn MG, Markofski MM, Pence BD, Hannemann RE. Adding exercise training to rosuvastatin treatment: influence on serum lipids and biomarkers of muscle and liver damage. *Metab Clin Exp* 2009;58:1030-38.
14. British Columbia Ministry of Health. Department of National Health and Welfare, Canada. Physical Activity Readiness Questionnaire – PAR-Q. Revised 2002, Canadian Society for Exercise Physiology. Available from: <http://www.csep.ca/forms>

15. Dwyer GB, Davis SE, editors. ACSM's Health-Related Physical Fitness Assessment Manual. Philadelphia: Lippincott Williams and Wilkins. 2005, p 43-171.
16. Durnin JVGA, Womersley J. Body fat assessment from total body density and its estimation from skinfold thickness: measurements on 481 men and women aged from 16-72 years. *Br J Nutr* 1974;32:77-97.
17. Dimsdale JE, Ziegler MG. What do plasma and urinary measures of catecholamines tell us about human response to stressors? *Circulation* 1991;83(Suppl. II):II-36-II-42.
18. Frayn KN. Calculation of substrate oxidation rates in vivo from gaseous exchange. *J Appl Physiol* 1983;55:628-34.
19. Faul F, Erdfelder E, Lang A-G, Buchner A. G*Power 3. A flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behav Res Methods* 2007;39:175-91.
20. Smolin LA, Grosvenor MB. Nutrition. Science and Applications. 3rd ed. Orlando: Harcourt College Publishing. 2000, p 92-197.
21. Crandall JP, Mather K, Rajpathak SN, Golberg RB, Watson K, Foo S, et al. Satin use and risk of developing diabetes: results from the Diabetes Prevention Program. *BMJ Open Diab Res Care* 2017;5:e000438doi:10.1136/bmjdr-2017-000438
22. Reents S. Antilipemic agents. In: Sport and Exercise Pharmacology. Champaign, IL: Human Kinetics. 2000, p 217-34.
23. Kreisberg RA, Oberman A. Medical management of Hyperlipidemia/Dyslipidemia. *J Clin Endocrinol Metab* 2003;88:2445-61.
24. Staels B, Dallongeville J, Auwerx J, Schoonjans K, Leitersdorf E, Fruchart J-C. Mechanism of action of fibrates on lipid and lipoprotein metabolism. *Circulation* 1998;98:2088-93.
25. Khan T, Hamilton MP, Mundy DI, Chua SC, Scherer PE. Impact of Simvastatin on adipose tissue: pleiotropic effects in vivo. *Endocrinology* 2009;150:5262-72.
26. Apostolopoulou M, Corsini A, Roden M. The role of mitochondria in statin-induced myopathy. *Eur J Clin Invest* 2015;45:745-54.
27. Golomb BA, Evans MA. Statin Adverse Effects: A review of the literature and evidence for a mitochondrial mechanism. *Am J Cardiovasc Drugs* 2008;8:373-418.
28. Laaksonen R, Jokelainen K, Laakso J, Sahi J, Härkönen M, Tikkanen MJ, et al. The effect of simvastatin treatment on natural antioxidants in low-density lipoproteins and high-energy phosphates and ubiquinone in skeletal muscle. *Am J Cardiol* 1996;77:851-54.
29. Marcoff L, Thompson PD. The role of coenzyme Q10 in statin-associated myopathy: a systematic review. *J Am Coll Cardiol* 2007;49:2231-37.

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30. Banach M, Serban C, Sahebkar A, Ursoniu S, Rysz, J, Muntner P, et al. Effects of coenzyme Q10 on statin-induced myopathy: a meta-analysis of randomized controlled trials. Lipid and Blood Pressure meta-analysis collaboration group. *Mayo Clin Proc* 2015;90:24-34.

31. Paolisso G, Barbagallo M, Petrella G, Ragno E, Barbieri M, Giordano M, et al. Effects of simvastatin and atorvastatin administration on insulin resistance and respiratory quotient in aged dyslipidemic non-insulin dependent diabetic patients. *Atherosclerosis* 2000;150:121-27.

32. Mikus CR, Boyle LJ, Borengasser SJ, Oberlin DJ, Naples SP, Fletcher J, et al. Simvastatin impairs exercise training adaptations. *J Am Coll Cardiol* 2013;62:709-14.

33. Martin WH, Klein S. Use of endogenous carbohydrate and fat as fuels during exercise. *Proc Nutr Soc* 1998;57:49-54.

34. O'Keefe JH, Schnohr P, Lavie CJ. The dose of running that best confers longevity. *Heart* 2013;99:588-90.

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455 **Table 1. Patient characteristics.**
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Variable	
N	16
Gender	13M, 3F
Age (years)	57 ±3
RHR (mmHg)	70 ±3
SBP (mmHg)	125 ±2
DBP (mmHg)	80 ±2
MAP (mmHg)	94 ±2
Height (cm)	173 ±2
Weight (kg)	84 ±3
BMI (kg/m ²)	28 ±1
Waist (cm)	95 ±3
Hip (cm)	103 ±2
Waist-hip ratio	0.91 ±0.02
%BF (skinfolds)	33 ±2
%BF (Tanita)	27 ±2
VO _{2max} (ml.kg ⁻¹ .min ⁻¹)	33 ±3

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458 N:number of participants; M:male; F:female; RHR:resting heart rate; SBP:systolic blood pressure;
459 DBP:diastolic blood pressure; MAP:mean arterial pressure; BMI:body mass index; %BF:percent body
460 fat; VO_{2max}:maximal oxygen uptake. Values are mean ±SEM.
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484 **Table 2. Dietary intake.**
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	Energy or % Macronutrient intake
Total energy (kJ/day)	10,111 ±927
Carbohydrate (%)	48 ±2
Protein (%)	17 ±1
Fat (%)	28 ±2
Monounsaturated fat (%)	11 ±0.8
Polyunsaturated fat (%)	5 ±0.5
Saturated fat (%)	9 ±0.5
Alcohol (%)	7 ±2

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487 Values are mean ±SEM.
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520 **Table 3. Fasting blood lipids (mmol/L) in the absence(-) and presence(+) of a statin.**
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	-statin	+statin	<i>p</i>-value
Total cholesterol	5.76 ±0.31	4.11 ±0.14	.000
Triglyceride	1.69 ±0.41	1.14 ±0.15	.073
HDL-C	1.31 ±0.10	1.28 ±0.10	.908
LDL-C	3.77 ±0.21	2.32 ±0.10	.000
TC/HDL	4.54 ±0.40	3.39 ±0.20	.001

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 523 HDL-C:high density lipoprotein cholesterol; LDL-C:low density lipoprotein cholesterol; TC/HDL:total
 524 cholesterol/high density lipoprotein ratio. Values are mean ±SEM. $p \leq 0.05$ significantly different between
 525 ±statin (Student's paired t-test).

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526 **Table 4. Physiological variable measured in participants during a walk at time = 15, 30 and 45**
527 **minutes in either the absence(-) or presence(+) of a statin.**
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Variable	-statin			+statin			p-value
	15 min	30 min	45 min	15 min	30 min	45 min	
Fat oxidation (g/min)	0.16±0.02	0.23±0.01	0.29±0.02	0.13±0.02	0.19±0.02	0.27±0.02	.622
CHO oxidation (g/min)	1.8±0.15	1.5±0.13	1.3±0.13	1.7±0.15	1.5±0.13	1.3±0.11	.312
%Fat	19±2	31±2	38±2	19±3	28±2	38±2	.567
%CHO	82±2	70±2	63±2	82±3	74±3	63±2	.390
RER	.93±007	.90±005	.88±006	.93±009	.91±009	.88±007	.393
Glycerol (µmol/l) (Cayman)	175±24	202±20	232±22	159±15	167±15	180±13	.080
Glycerol (µmol/l) (Sigma)	102±8	125±11	150±15	99±15	105±14	125±13	.178
FFA (µmol/L)	47±8	82±12	138±22	53±13	70±16	104±20	.033
VO ₂ (ml.kg ⁻¹ .min ⁻¹)	16±1.2	15±1.3	15±1.3	15±1.2	15±1.3	15±1.3	.452
Glucose (mmol/l)	4.2±0.2	4.0±0.2	4.2±0.1	4.3±0.3	4.2±0.2	4.3±0.1	.883
Lactate (mmol/l)	1.3±09	1.0±06	1.0±09	1.2±08	1.0±06	0.9±03	.426
HR (bpm)	109±3	110±3	111±3	101±3	103±3	104±3	.756
RPE	11±0.4	11±0.4	12±0.4	11±0.4	11±0.4	11±0.4	.596

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530 CHO:carbohydrate; %Fat:percentage kcal derived from fat; %CHO:percentage kcal derived from
531 carbohydrate; RER:respiratory exchange ratio; FFA:free fatty acid; VO₂:oxygen uptake; HR(bpm):heart
532 rate (beats per minute); RPE:rating of perceived exertion. Data are mean±SEM. Significance is P≤ 0.05,
533 repeated measures analysis of variance (ANOVA). No significance achieved for 'treatment and time' for
534 any variables (exception FFA). For FFA, significance is p≤0.016 (Bonferroni adjustment) with the
535 simple main effect also not statistically significant.