

Daily Step Count and Postprandial Fat Metabolism

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ABSTRACT

BURTON, H. M., and E. F. COYLE. Daily Step Count and Postprandial Fat Metabolism. *Med. Sci. Sports Exerc.*, Vol. 53, No. 2, pp. 333–340, 2021. **Introduction:** Two benefits of acute exercise are the next day's lowering of the postprandial plasma triglyceride response to a high-fat meal and increased fat oxidation. However, if activity levels (daily steps) are very low, these acute adaptations to exercise do not occur. This phenomenon has been termed "exercise resistance." This study sought to systematically reduce daily step number and identify the range of step counts that elicit "exercise resistance." **Methods:** Ten participants completed three, 5-d trials in a randomized, crossover design with differing levels of step reduction. After 2 d of controlled activity, participants completed 2 d of LOW, LIMITED, or NORMAL steps (2675 ± 314 , 4759 ± 276 , and 8481 ± 581 steps per day, respectively). Participants completed a 1-h bout of running on the evening of the second day. High-fat tolerance tests were performed on the next morning, and postprandial responses were compared. **Results:** After LOW and LIMITED, postprandial incremental area under the curve (AUC) of plasma triglyceride was elevated 22%–23% compared with NORMAL ($P < 0.05$). Whole body fat oxidation was also significantly lower (16%–19%, $P < 0.05$, respectively) in LOW and LIMITED compared with NORMAL. No significant differences were found between LOW and LIMITED. **Conclusion:** Two days of step reduction to approximately 2500–5000 steps per day in young healthy individuals impairs the ability of an acute bout of exercise to increase fat oxidation and attenuate postprandial increases in plasma triglycerides. This suggests that "exercise resistance" occurs in individuals taking approximately 5000 or fewer steps per day, whereas 8500 steps per day protects against exercise resistance in fat metabolism. It seems that fat metabolism is influenced more by the inhibitory effects of inactivity than by the stimulating effects derived from 1 h of moderate-intensity running. **Key Words:** INACTIVITY, EXERCISE, HEALTH, STEP REDUCTION, POSTPRANDIAL LIPEMIA, WALKING, LIPOPROTEIN LIPASE, SITTING

The cardiometabolic health benefits of physical activity and exercise such as improved postprandial hypertriglyceridemia and improved glucose tolerance can be gained acutely from a single bout of exercise and lost with several days of inactivity (1–3). However, in studies in which acute exercise resulted in reduced and improved postprandial hypertriglyceridemia, the participants were accumulating approximately 7000–8500 steps on the day before evaluation of postprandial metabolism (4–6). At the low end of daily steps, Kim et al. (7) reported that in participants who were sitting for $>14 \text{ h} \cdot \text{d}^{-1}$ and taking only 1650 steps per day, a

1-h bout of running at 67% maximal oxygen consumption ($\dot{V}O_{2\text{max}}$) failed to improve postprandial hypertriglyceridemia the next morning. It seems that physical inactivity (i.e., high sitting and severely reduced step count) rendered the participants resistant to the normal acute improvements in indices of cardiometabolic health that are normally derived from a 1-h bout of running. This phenomenon is called "exercise resistance" (7). A follow-up study in 2019 (8), using additional controls to verify the existence of this phenomenon, found that a group taking ~ 3700 steps per day also exhibited exercise resistance. Therefore, it is important to systematically delineate what level of daily steps causes impairment of the ability of acute exercise to improve the postprandial plasma triglyceride response and fat oxidation.

In modern culture, we have engineered physical activity out of our daily lives. Periods of prolonged inactivity, characterized by mostly sitting, have become routine in the lives of many and coincide with the nonfasting or postprandial state. In the postprandial state, triglyceride levels in the plasma can remain elevated for up to 10 h, typically peaking 3–6 h after a meal rich in fat (9). The magnitude and duration of this elevation is influenced by previous physical activity (10–12), diet

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(13), and genetics (14,15). As demonstrated in epidemiological studies (16,17), nonfasting plasma triglyceride levels, i.e., postprandial lipemia (PPL), better predict cardiovascular events than fasting plasma triglyceride levels and are known to be associated with diseases, including metabolic syndrome, type 2 diabetes, and atherosclerosis. In fact, in several recent epidemiological studies, inactivity and/or sitting time have been strongly associated with the risk of obesity, metabolic disorders including type 2 diabetes mellitus, and especially with cardiovascular disease (CVD) and death (18–20). Surprisingly, some have reported that the risks from prolonged sitting seem “independent” of the volume of exercise being performed (18–21). This means people who meet the recommended guidelines (AHA or American College of Sports Medicine [ACSM]) for physical activity of 150 min·wk⁻¹ of moderate-intensity exercise seem to still be at risk for developing CVD and all-cause death if they have a lifestyle routinely incorporating prolonged periods of inactive, sedentary behavior (>10–12 h·d⁻¹) and low steps per day.

One of the strongest inverse correlations with sedentary time is time in light-intensity activity, such as walking (22). This means of course that increasing time spent ambulatory reduces sitting time. Accordingly, the manipulation of daily step count has been a popular and practical method of quantifying inactivity. Appropriately, increased daily walking has been shown to reduce cardiovascular events (23), whereas reductions in daily step number for as little as 1 wk have been associated with drastic increases in the area under the curve of plasma insulin during an oral glucose tolerance test (24). This increase showed the potential to grow to nearly 80% greater, if the reductions were maintained for two more weeks (24). Daily step reductions have also been linked to decreased $\dot{V}O_{2max}$, endothelial dysfunction, decreased insulin sensitivity, decreased lean leg mass, and increased abdominal fat (24–26).

Meanwhile, it is well established that, in the absence of high levels of daily sitting, a single bout of moderate exercise lasting 60–90 min attenuates PPL regardless of previous lipid levels (12) and training status (10,27,28). Many studies (10–12) have been conducted to investigate the effect of a single bout of moderate-intensity exercise on postprandial triglyceride

levels in comparison to a control condition. Participants in these studies were asked to refrain from any planned exercise but their ambulatory activity, and walking was not carefully controlled. Furthermore, very few studies (7,8) have systematically investigated the collective effects of daily step reduction combined with moderate acute exercise. Recent studies (7,8) that drastically reduced daily step number abolished the ability of an acute bout of exercise to increase fat oxidation the next day and attenuate the increase in PPL. Some other studies seem to suggest that exercise resistance may not occur when taking as little as ~7900 steps per day (4).

Thus, the purpose of this study was to systematically investigate the effect of reductions in daily step number and a single 1-h bout of moderate-intensity exercise to improve the next day’s postprandial responses of plasma triglyceride and glucose, as well as fat oxidation. We hypothesized that postprandial responses, after a single bout of 1-h of running at ~65% $\dot{V}O_{2max}$ performed the evening before, would be impaired progressively as daily steps decreased.

METHODS

Ten healthy, recreationally active male ($n = 7$) and female ($n = 3$) participants completed three trials of differing daily step counts based on previously established benchmarks for daily physical activity (29). Participants were asked to take approximately 2500, 5000, and 8500 steps per day for low activity (LOW), limited activity (LIMITED), and what is considered normal activity (NORMAL), respectively, in a crossover design, each occurring over 5 d with at least a week interval between trials (Fig. 1). Participants were asked to refrain from any planned exercise outside of the experimental design. Participants were given written and verbal description of all the procedures and measurements used in this study, and written informed consent was obtained. The Institutional Review Board of the University of Texas at Austin approved this study (ClinicalTrials.gov Identifier: NCT03697382).

Experimental design. Each trial consisted of three phases (Fig. 1). The first 2 d served as a control phase (C1 and C2),

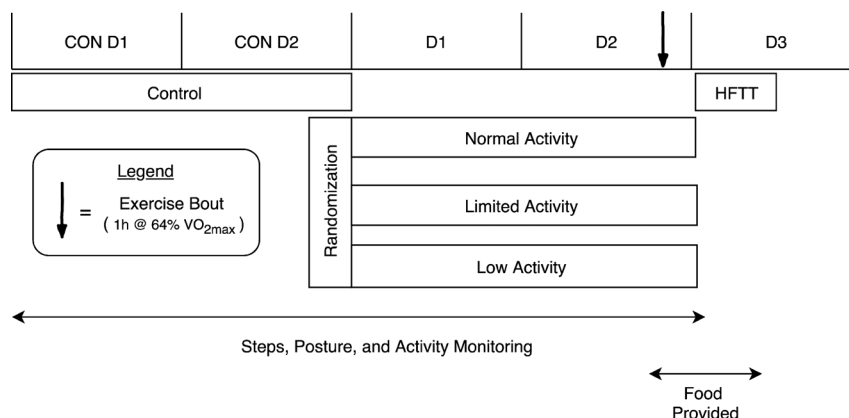


FIGURE 1—Study design. Participants completed a 5-d randomized, crossover experimental design with differing levels of daily step reduction (i.e., Low, 2675; Limited, 4759; and Normal activity, 8481 steps per day). Participants completed two control days with activity monitoring before the initiation the 2-d step reduction (D1 and D2). Participants also completed an hour of treadmill running on the night of D2 followed by HFTT on the morning of D3.

which allowed for familiarization with measurements, followed by a 2-d intervention phase consisting of LOW, LIMITED, or NORMAL physical activity consisting of daily step counts that amounted to 2675 ± 314 , 4759 ± 276 , or 8481 ± 581 steps per day, respectively. On the evening of the second day of each trial, subjects ran for 1 h at 64% $\dot{V}O_{2\max}$ on a laboratory treadmill. On the morning of day 3, after the previous evenings run, all participants ingested a high-fat shake (i.e., high-fat tolerance tests [HFTT]), and the postprandial responses were measured over the subsequent 6-h period. Throughout the three trials, participants were instructed to refrain from any exercise other than that prescribed in the study design. Participants were also asked to keep a consistent sleep/wake cycle during the trials.

Preliminary testing. One week before the initiation of the first trial, participants visited the Human Performance Laboratory for a 20-min, four-stage submaximal test to determine oxygen consumption while jogging at different paces followed by determination of maximal oxygen uptake ($\dot{V}O_{2\max}$). This served to determine the appropriate treadmill speed to elicit the desired intensity during the 1-h exercise bout. To determine $\dot{V}O_{2\max}$, participants performed an incremental treadmill test lasting 8–12 min during which the incline was increased 2% every 2 min (30). $\dot{V}O_2$, $\dot{V}CO_2$, and heart rate were monitored throughout the test, and the highest 30-s $\dot{V}O_2$ average was recorded for the participant's maximal oxygen consumption. The ACSM criteria for $\dot{V}O_{2\max}$ were used in assessing a successful $\dot{V}O_{2\max}$ test.

Control phase. Participants were instrumented with an activity monitor worn on the mid thigh to record step count (activPAL; PAL Technologies, Glasgow, Scotland), and the monitor began recording at 0000 h on the first day of the control phase (C1). Participants were asked to remain aware of their step count and to limit steps to approximately 10,000 or less during the control period (31). The activPAL provided an objective measure of daily steps throughout the duration of each trial. Visual feedback for daily steps is unavailable on the activPAL; therefore, participants were asked to use a second pedometer worn on the hip or wrist or an enabled smart phone to provide feedback on daily steps.

Intervention phase. During the intervention phase, D1 and D2, participants were asked to remain seated or lying for much of the day to accommodate their assigned level of nonexercise activity (2675, 4759, or 8481 steps per day). On D2 of each trial, participants continued to adhere to the assigned step count and additionally completed a 1-h run at 64.4% $\dot{V}O_{2\max}$ at 1800 h. The steps during this bout of exercise were not included as part of the participants step total for D2.

HFTT. On the day of the HFTT (D5), participants reported to the laboratory at 0700 h. Body weight was measured. They then lie for 5 min before insertion of a catheter into an antecubital vein. A fasting blood sample was obtained and fat oxidation was measured before consumption of a high-fat shake (mostly melted ice cream and heavy cream, approximately $14.8 \text{ kcal}\cdot\text{kg}^{-1}$ [0.8 g, 1.2 g, and 0.2 $\text{g}\cdot\text{kg}^{-1}$ BW of carbohydrate, fat, and protein, respectively]). Blood samples were collected over the next 6 h at 2, 3, 4 and 6 h postconsumption of the high-fat shake. All

blood samples collected were transferred to K_2 EDTA collection tubes (BD), centrifuged at 2000g for 15 min at 4°C and then stored in –80°C freezer until later analysis. During HFTT, participants were asked to remain seated quietly reading, watching movies, and/or surfing the Internet. Participants were allowed to use the restroom.

Postprandial substrate oxidation. Pre- and postprandial expired gas collection was used to assess substrate oxidation. Participants rested in a chair for 10 min, followed by expired gas collection through meteorological balloons for 10 min at 0, 2, 4, and 6 h. It has been previously demonstrated that inactivity reduces whole body fat oxidation (7).

Energy expenditure and substrate oxidation were calculated from oxygen consumption, carbon dioxide production, and RER; energy expenditure and substrate oxidation were calculated based on the methods of Frayn (32).

$$\begin{aligned} \text{\% energy from carbohydrate (CHO) oxidation} \\ = ((\text{RER} - 0.707)/0.293) \times 100 \end{aligned}$$

$$\text{\% energy from fat oxidation} = 100 - \text{\% energy from CHO oxidation}$$

$$\begin{aligned} \text{CHO oxidation (kcal}\cdot\text{min}^{-1}) \\ = [(\text{\%CHO oxidation}/100) \times \dot{V}O_2] \times 5.05 \text{ kcal}\cdot\text{L}^{-1} \text{ O}_2 \end{aligned}$$

$$\begin{aligned} \text{fat oxidation (kcal}\cdot\text{min}^{-1}) = [(1 - \text{\%CHO oxidation}/100) \\ \times \dot{V}O_2] \times 4.7 \text{ kcal}\cdot\text{L}^{-1} \text{ O}_2 \end{aligned}$$

$$\text{energy expenditure (kcal}\cdot\text{min}^{-1}) = \text{CHO oxidation} + \text{fat oxidation}$$

Dietary control. During the study, participants were asked to eat to satiety. Participants logged all food and were asked to consume the same foods on the days before each HFTT. On the evening before the HFTT, participants were given a low-fat meal to consume as fat in the previous meal can affect the response to a high-fat test meal (6,33). Participants were allowed to supplement higher energy expenditure during the LIMITED and the NORMAL step trials with a small snack but were asked to adhere to a diet standard in macronutrient breakdown (34).

Biochemical analysis. For plasma triglyceride and glucose concentrations, all blood samples collected were immediately transferred to K_2 EDTA collection tubes (BD Vacutainer, Franklin Lakes, NJ), centrifuged at 3000g for 15 min at 4°C. Plasma was then stored in separate aliquots at –80°C until later analysis. All measurements for each participant were performed in duplicate within the same analysis. Plasma triglyceride and glucose concentrations were measured by a spectrophotometric method using commercially available kits (Pointe Scientific, Inc., Canton, MI). Intra-assay coefficients of variation for plasma triglyceride and glucose concentrations were all less than 10%.

Statistical analysis. Incremental (AUC_I) and total area under the curve (AUC_T) for plasma triglyceride and glucose were calculated. Once calculated, repeated-measures one-way ANOVA was used to test for differences. Plasma glucose and triglyceride curves were calculated and analyzed using repeated-measures two-way ANOVA (trial \times time). Daily step counts were analyzed using repeated-measures two-way ANOVA (trial \times time). Similarly, RER as well as fat and carbohydrate oxidation were analyzed using repeated-measure two-way ANOVA (trial \times time). When interactions were significant, Tukey's

TABLE 1. Average daily steps measured via activPAL activity monitor, attached on the participant's anterior thigh throughout each trial.

Daily Steps	Trial	Day of Trial			
		C1	C2	D1	D2
	Low	10,198 ± 1476	11,015 ± 1159	2744 ± 331	2605 ± 313
	Limited	9808 ± 1207	10,568 ± 1112	4482 ± 318*	5037 ± 206**
	Normal	11,056 ± 1324	10,732 ± 936	8431 ± 732†	8530 ± 420†

Data are presented as mean ± SE. Average daily step counts for each trial are presented for control (C1 and C2) and intervention phases (D1 and D2).

*Significantly different from Low, $P < 0.05$.

**Significantly different from Low, $P < 0.01$.

†Significantly different from both Low and Limited, $P < 0.01$.

honestly significant difference *post hoc* tests were run. All data were analyzed using GraphPad Prism 8 (GraphPad Software Inc., La Jolla, CA). All data are expressed as mean ± SE, unless otherwise noted; the level for statistical significance was set at $P \leq 0.05$.

RESULTS

Participant characteristics. Ten participants (seven males and three females) were recruited from the Austin area. These participants were young, healthy, untrained to recreationally active individuals (age = 25.7 ± 1.8 yr, body mass = 79.7 ± 5.4 kg, height = 173 ± 3 cm, BMI = 26.4 ± 1.2 kg·m⁻², $\dot{V}O_{2\max} = 42.7 \pm 1.4$ mL·kg⁻¹·min⁻¹).

Responses to submaximal exercise. Submaximal jogging for 1 h elicited a heart rate of 154 ± 4 bpm, an oxygen consumption that equated to 64.4% of $\dot{V}O_{2\max}$, and participants reported an RPE of 11.4 ± 1 . All of these are indicative of moderate-intensity exercise.

Daily steps. Daily steps are presented in Table 1. No significant differences were noted within or between trials for control days, which averaged $10,648 \pm 824$ steps per day ($P > 0.05$). However, daily steps averaged on days 1 and 2 of the intervention, excluding exercise steps on day 2, were significantly different for all trials (LOW, 2675 ± 314 ; LIMITED, 4759 ± 276 ; NORMAL, 8481 ± 581 ; $P < 0.01$).

Plasma triglyceride responses. The postprandial plasma triglyceride concentration responses are shown in Figure 2, with LOW and LIMITED being significantly higher than NORMAL at 2, 3, and 4 h ($P < 0.05$). Incremental area under the curve (AUC_I) in LOW and LIMITED were elevated by 22%–23% ($P < 0.05$) compared with NORMAL (Fig. 3). Total area under the curve for plasma triglyceride (AUC_T) is shown in Figure 3, with LOW being significantly higher than NORMAL ($P < 0.01$) and a trend toward LIMITED being higher than NORMAL ($P = 0.09$). No differences were detected for postprandial plasma triglyceride concentration between LOW and LIMITED in AUC_T and AUC_I.

Plasma glucose concentrations. Plasma glucose concentration is shown in Figure 4. Plasma glucose concentration as well as areas under the curve showed no differences ($P > 0.05$) between the three trials in AUC_T or AUC_I.

Postprandial substrate oxidation. Postprandial substrate RER was significantly higher than NORMAL (0.77 ± 0.01) and LIMITED (0.80 ± 0.01 , $P < 0.05$), with a strong trend toward significant from LOW (0.81 ± 0.01 , $P = 0.06$; Table 2). Absolute fat oxidation was significantly lower than NORMAL (396 ± 27 kcal), with LOW (319 ± 34 kcal; -19% , $P < 0.05$) and LIMITED (342 ± 30 kcal, -14% , $P < 0.05$).

DISCUSSION

The purpose of this study was to investigate the effect of daily step reductions on postprandial responses to a high-fat meal consumed the morning after an acute bout of moderate-intensity exercise. The primary finding was that when individuals took only 2675 or 4759 daily steps (LOW and LIMITED), their postprandial plasma triglyceride responses and whole body fat oxidation were significantly impaired compared with when taking 8431 daily steps (NORMAL), although it should be noted that their freely chosen steps in the control phase were approximately 10,000 steps per day. Participants displayed a

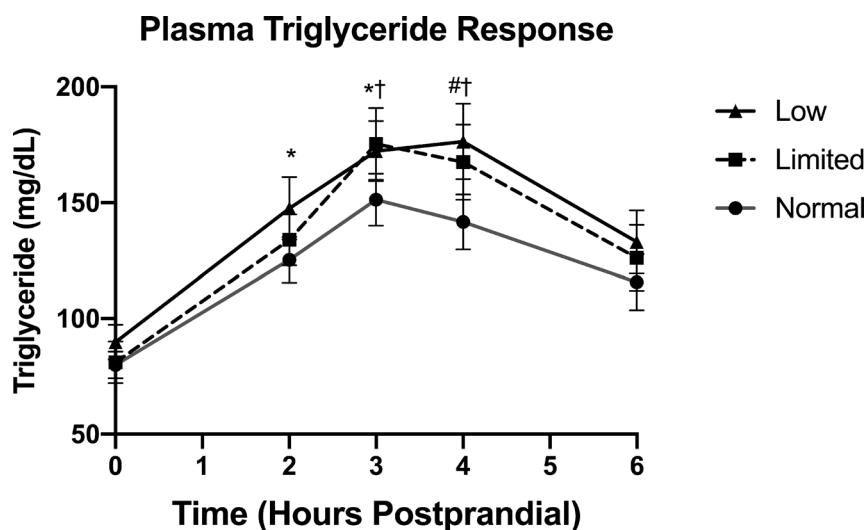
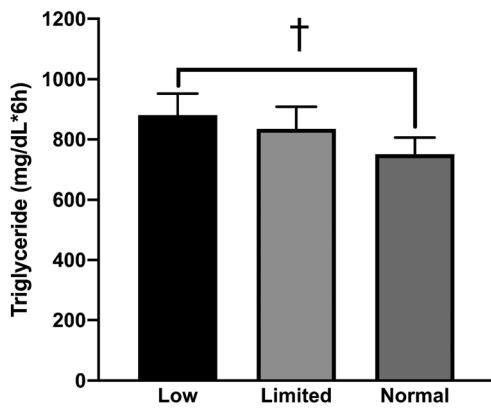


FIGURE 2—Temporal responses of plasma triglyceride concentrations for each trial during HFTT. *Normal significantly different from Low, $P < 0.05$. †Normal significantly different from Limited, $P < 0.05$. #Normal significantly different from Low, $P < 0.01$. Data are reported as mean ± SE.

Plasma Triglyceride Total Area Under the Curve



Plasma Triglyceride Incremental Area Under the Curve

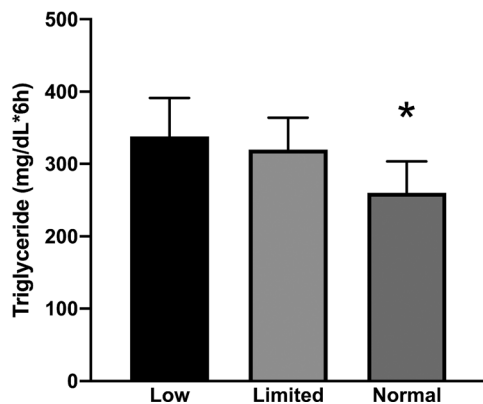


FIGURE 3—Total and incremental areas under the curve of plasma triglyceride concentrations during HFTT for each trial. *Significantly different from Low and Limited step group, $P < 0.05$. †Significantly different from Low step group, $P < 0.01$. Data are reported as mean \pm SE.

14%–19% reduction in fat oxidation and a concomitant 22%–23% increase in TG AUC₁ when taking 2675–4759 steps per day compared with 8481 steps per day. Despite completing identical and substantial 1-h bouts of running (64% $\dot{V}O_{2max}$) the night before the HFTT, participants still displayed decreased

responsiveness to this exercise if they reduced daily steps to approximately 2500–5000 steps per day, at least in regard to whole body fat oxidation and PPL. The increase in plasma TG concentration above NORMAL during LOW and LIMITED may be due to a reduced uptake of plasma triglyceride by tissue and reduced fat oxidation in LOW and LIMITED compared with NORMAL.

The 22%–23% increase in TG AUC₁ above NORMAL and the concomitant 14%–19% reduction in fat oxidation below NORMAL with LOW and LIMITED are generally similar to the 20%–40% differences in TG AUC₁ reported previously for nonexercising controls compared with an exercise treatment (4,5,12,35). This lends credence to the emerging ideas concerning the interaction of physical inactivity and exercise (1,24). Although a 1-h bout of moderate-intensity running was performed the evening before the evaluation of fat metabolism in all trials, the effects of the previous days' low physical activity (i.e., LOW and LIMITED) were carried forward and impaired fat metabolism. This suggests that postprandial fat metabolism is influenced more by the inhibitory effects of inactivity than by the stimulating effects of exercise, at least with the current design.

Our findings are interesting in light of recent findings of a phenomenon termed “exercise resistance” (7,8). We previously observed that individuals taking less than 4000 daily steps were resistant to the exercise stimulus provided by 1-h of running at $\sim 65\%$ $\dot{V}O_{2max}$, in terms of improving postprandial fat metabolism the morning after exercise. In these randomized crossover trials (7,8,36), the protective effects of exercise in preventing exaggerated rises in postprandial plasma triglycerides were not realized if daily step counts were reduced to <4000 steps per day by imposed sitting for 13 h. By imposing high required daily sitting, these investigators considerably reduced the amount of muscular activity of their participants as light-intensity activity makes up the majority of daily energy expenditure (17). It seems that by drastically reducing the contractile activity in the study participants, an environment was produced that prevented the classic improvement of the next day's responses to the exercise stimulus. It has been postulated in a recent meta-analysis by Ekelund et al. (37) that

Plasma Glucose Response

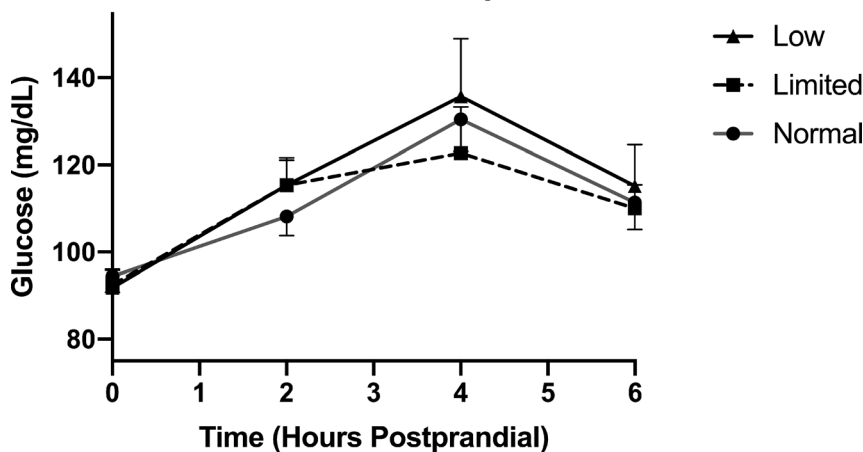


FIGURE 4—Temporal responses of plasma glucose concentrations for each trial during HFTT. Data are reported as mean \pm SE.

TABLE 2. Overall postprandial substrate oxidation during HFTT for each trial.

Variables	Treatment Group		
	Low	Limited	Normal
RER	0.81 ± 0.01	0.80 ± 0.01	0.77 ± 0.01*
Fat oxidation (% energy)	66.1 ± 4.87	69.6 ± 3.93	80.4 ± 2.65*
Fat oxidation (kcal/6 h)	318.9 ± 34.5	342.4 ± 30.9	396.0 ± 27.5*
CHO oxidation (% energy)	33.9 ± 4.87	30.4 ± 3.93	19.6 ± 2.65*
CHO oxidation (kcal/6 h)	164.0 ± 25.3	149.0 ± 24.1	97.8 ± 12.8*
Total energy expenditure (kcal/6 h)	482.9 ± 32.6	491.4 ± 31.2	493.8 ± 27.0

Data are reported as mean ± SE.

*Significantly different from Low and Limited, $P < 0.05$.

individuals experiencing high levels of daily inactivity are at an increased risk of mortality even when participating in moderately high levels of daily activity or exercise. It seems inactivity may exert deleterious effects on the muscle beyond fat metabolism as well. Recent observations support this contention in that not only is PPL impaired but reduced myofibrillar protein synthesis can be seen in elderly (26) and young healthy individuals (38) in response to step reductions to ~1400 steps per day over 2 wk or less. Taken together, it seems that reduced contractile activity for long periods of the day causes a condition in which current exercise recommendations may not be effective for deriving some protective health benefits of exercise such as improved fat oxidation and PPL responses. In populations regularly experiencing prolonged sitting and inactivity (i.e., <5000 steps per day), it is not known if or what type of exercise regime might overcome “exercise resistance” in terms of reversing impaired fat metabolism, given the present observation that 1-h of running the evening before was ineffective. At least in terms of fat metabolism, it seems that approximately 8500 steps per day are needed to prevent exercise resistance. Previous studies suggest that avoiding exercise resistance may occur with as little as ~7900 steps per day (4). We have recently observed that if prolonged sitting throughout the day is interrupted hourly by 20 s of maximal cycling (5 × 4-s bouts), exercise resistance can be prevented at least in terms of fat metabolism (39). It thus seems that exercise resistance can be prevented not just by accumulating steps throughout the day but also by accumulating bouts of very short maximal-intensity exercise. This implies that accumulating at least 8500 steps per day, although effective, is not the only method for preventing exercise resistance of fat metabolism.

Recent research has begun to place a particular emphasis on the benefits of increases in daily step counts given the ease of measurement and application (31,40–42). Some have recently proposed an inverse dose–response relationship between daily step counts and markers of healthy metabolism. Olsen et al. (24) found that reducing steps from ~10,500 to ~1400 steps per day for 2 wk increased postprandial TG AUC_T by 21% in the absence of exercise. Others have found that the incidence of CVD, type 2 diabetes, and all-cause mortality decreases when daily step count is increased (41) (42). Lee et al. (42) found rates of mortality progressively declined with increasing daily steps until plateauing at approximately 7500 steps per day. Contrary to the findings in the present study, the authors reported that groups taking as few as 4363 steps per day displayed reduced mortality compared with those

taking 2718 step per day. The difference in findings may be due to several factors, foremost, the different end points (i.e., acute fat metabolism vs mortality). Furthermore, the participants in Lee et al. (42) were substantially older than the population recruited for the current study. Moreover, a recent meta-analysis (41) suggested a 10% reduced risk of cardiovascular events for each 2000-step increase in daily step number up to 10,000 steps per day. These investigations differ from the findings in this study, as we did not find any improvements in acute postprandial plasma triglyceride responses or fat oxidation when an individual increased daily walking from ~2500 to ~5000 steps per day. A few important distinctions should be noted and may explain this discrepancy. First, and most obviously, the current investigation focused on responses the morning after moderate exercise and, thus, the benefits of acute exercise on fat metabolism, which were not used in the aforementioned studies. Second, the “baseline” step counts in almost all of these investigations exceeded 5000 steps per day (41–43), which excludes comparisons below this level of daily walking, such as the 2675 step per day trial presented herein. The current investigation was also conducted over a much shorter period than the observations with different end points. Lastly, PPL, although indicative of CVD events, is only a single factor contributing to the development of CVD and should not be considered equivalent or wholly indicative of CVD.

Daily step count at or below 5000 should be classified as a “sedentary lifestyle index” (31,40) and should be viewed as problematic because of the distinct health ramifications seen below this level of activity due to “nonexercise activity deficiency” (44). Although this is a reduction below the level that some would consider normal, it should not be disregarded. In fact, estimates from the NHANES study, based on objectively collected accelerometer data, indicate approximately 37% of the U.S. population would fall below this 5000 steps per day threshold of physical activity (29). Moreover, technological advances have drastically decreased occupational physical activity such that activity in much of the workforce can approximate that of sedentary, elderly individuals (45). The occurrence of these high levels of inactivity (i.e., <5000 steps per day) is not obviated completely even in health-conscious individuals. For example, “workday” sedentary time in 208 marathon and half-marathon participants was observed to be similar to those seen in the elderly in assisted living communities (46,47). When making recommendation regarding the steps per day count needed to elicit a normal postprandial fat metabolism, it was observed that approximately 8500 was effective, but 5000 and 2500 steps per day were not effective. Therefore, the lowest step count that we can recommend based on the current design is 8500 steps per day. However, we do not know about the effectiveness of any step count in the range of 5000–8500 steps per day.

Muscle lipoprotein lipase (LPL) is the rate-limiting enzyme for the clearance of plasma triglycerides (48). Therefore, decreased LPL activity is a logical candidate for explaining the increased PPL found in this study with LOW and LIMITED.

Although not measured in the present investigation, low levels of contractile activity have been observed to drastically reduce the activity in LPL in muscle (49). In an animal model, hindlimb immobilization has been shown to have sizeable reductions (i.e., 90% decrease) in LPL activity, which developed rapidly (>60% reduction in <12 h) (49). However, it seems the downregulation is posttranscriptional. Even in the tissue with more than 90% reduction in LPL activity, LPL mRNA was similar to baseline (49). In addition, the data from Bey and Hamilton (49) suggest that LPL mRNA is not increased with walking or levels of contractile activity associated with maximal increases in LPL activity. It might be that LPL activity is downregulated by some metabolite (e.g., GPIHBP1 protein endocytosis) induced during periods of prolonged inactivity (50,51). GPIHBP1 is the protein responsible for anchoring LPL to the luminal surface of the capillary, and impairing proper translocation could suppress LPL activity without significant reductions in transcription (51). However, this hypothesis was not tested in the present study and is introduced only as a possible example.

In conclusion, to the best of our knowledge, the current investigation is the first to indicate that 2 d of step reductions to

approximately 2500–5000 steps per day can decrease an individual's responsiveness to an acute aerobic exercise bout in terms of stimulating the next day's improvements in fat oxidation and PPL. When participants took 5000 or fewer daily steps and despite performing a 1-h bout of exercise, they displayed a 16%–19% decrease in fat oxidation and a 22%–23% increase in postprandial plasma TG excursions the next day compared with NORMAL (approximately 8500 steps per day; $P < 0.05$). Therefore, it could be recommended that for optimal fat metabolism, people do not reduce their step count below approximately 8500 steps per day, even if they are additionally exercising at a moderate intensity.

The results of the study are presented clearly, honestly, and without fabrication, falsification, or inappropriate data manipulation. The results of the present study do not constitute endorsement by the ACSM. The authors declare no conflicts of interest in this unfunded research.

Author contributions: H. M. B. and E. F. C. conceived the research and designed the experiment; H. M. B. recruited subjects and performed experiments; H. M. B. and E. F. C. interpreted results of experiments; H. M. B. prepared figures, performed statistical analyses, and drafted the manuscript; H. M. B. and E. F. C. edited and revised manuscript; H. M. B. and E. F. C. approved final version of manuscript.

REFERENCES

- Holloszy J, Rennie M, Hickson R, Conlee R, Hagberg J. Physiological consequences of the biochemical adaptations to endurance exercise. *Ann N Y Acad Sci.* 1977;301(1):440–50.
- Holloszy JO, Kohrt WM. Regulation of carbohydrate and fat metabolism during and after exercise. *Annu Rev Nutr.* 1996;16:121–38.
- Holloszy JO, Skinner JS, Toro G, Cureton TK. Effects of a six month program of endurance exercise on the serum lipids of middle-age man. *Am J Cardiol.* 1964;14:753–60.
- Trombold JR, Christmas KM, Machin DR, Kim IY, Coyle EF. Acute high-intensity endurance exercise is more effective than moderate-intensity exercise for attenuation of postprandial triglyceride elevation. *J Appl Physiol (1985).* 2013;114(6):792–800.
- Kim IY, Park S, Trombold JR, Coyle EF. Effects of moderate- and intermittent low-intensity exercise on postprandial lipemia. *Med Sci Sports Exerc.* 2014;46(10):1882–90.
- Trombold JR, Christmas KM, Machin DR, et al. Postexercise macronutrient intake and subsequent postprandial triglyceride metabolism. *Med Sci Sports Exerc.* 2014;46(11):2099–106.
- Kim I-Y, Park S, Chou T-H, Trombold JR, Coyle EF. Prolonged sitting negatively affects the postprandial plasma triglyceride lowering effect of acute exercise. *Am J Physiol Endocrinol Metab.* 2016;311(5):E891–8.
- Akins JD, Crawford CK, Burton HM, Wolfe AS, Vardarli E, Coyle EF. Inactivity induces resistance to the metabolic benefits following acute exercise. *J Appl Physiol (1985).* 2019;126(4):1088–94.
- Patsch JR, Miesenböck G, Hopferwieser T, et al. Relation of triglyceride metabolism and coronary artery disease. Studies in the postprandial state. *Arterioscler Thromb Vasc Biol.* 1992;12(11):1336–45.
- Malkova D, Evans RD, Frayn KN, Humphreys SM, Jones PR, Hardman AE. Prior exercise and postprandial substrate extraction across the human leg. *Am J Physiol Endocrinol Metab.* 2000;279(5):E1020–8.
- Gill JM, Hardman AE. Postprandial lipemia: effects of exercise and restriction of energy intake compared. *Am J Clin Nutr.* 2000;71(2):465–71.
- Zhang JQ, Ji LL, Fretwell VS, Nunez G. Effect of exercise on postprandial lipemia in men with hypertriglyceridemia. *Eur J Appl Physiol.* 2006;98(6):575–82.
- Thomas TR, Fischer BA, Kist WB, Horner KE, Cox RH. Effects of exercise and n-3 fatty acids on postprandial lipemia. *J Appl Physiol.* 2000;88(6):2199–204.
- Tanaka T, Ordovas JM, Delgado-Lista J, et al. Peroxisome proliferator-activated receptor alpha polymorphisms and postprandial lipemia in healthy men. *J Lipid Res.* 2007;48(6):1402–8.
- Weiss EP, Brandauer J, Kulaputana O, et al. FABP2 Ala54Thr genotype is associated with glucoregulatory function and lipid oxidation after a high-fat meal in sedentary nondiabetic men and women. *Am J Clin Nutr.* 2007;85(1):102–8.
- Nordestgaard BG, Benn M, Schnohr P, Tybjaerg-Hansen A. Nonfasting triglycerides and risk of myocardial infarction, ischemic heart disease, and death in men and women. *JAMA.* 2007;298(3):299–308.
- Bansal S, Buring JE, Rifai N, Mora S, Sacks FM, Ridker PM. Fasting compared with nonfasting triglycerides and risk of cardiovascular events in women. *JAMA.* 2007;298(3):309–16.
- Biswas A, Oh PI, Faulkner GE, et al. Sedentary time and its association with risk for disease incidence, mortality, and hospitalization in adults: a systematic review and meta-analysis. *Ann Intern Med.* 2015;162(2):123–32.
- van der Ploeg HP, Chey T, Korda RJ, Banks E, Bauman A. Sitting time and all-cause mortality risk in 222 497 Australian adults. *Arch Intern Med.* 2012;172(6):494–500.
- Young DR, Hivert MF, Alhassan S, et al. Sedentary behavior and cardiovascular morbidity and mortality: a science advisory from the American Heart Association. *Circulation.* 2016;134(13):e262–79.
- Patel AV, Bernstein L, Deka A, et al. Leisure time spent sitting in relation to total mortality in a prospective cohort of US adults. *Am J Epidemiol.* 2010;172(4):419–29.
- Hamilton MT, Healy GN, Dunstan DW, Zderic TW, Owen N. Too little exercise and too much sitting: inactivity physiology and the need for new recommendations on sedentary behavior. *Curr Cardiovasc Risk Rep.* 2008;2(4):292–8.
- Manson JE, Greenland P, LaCroix AZ, et al. Walking compared with vigorous exercise for the prevention of cardiovascular events in women. *N Engl J Med.* 2002;347(10):716–25.
- Olsen RH, Krogh-Madsen R, Thomsen C, Booth FW, Pedersen BK. Metabolic responses to reduced daily steps in healthy nonexercising men. *JAMA.* 2008;299(11):1261–3.

25. Krogh-Madsen R, Thyfault JP, Broholm C, et al. A 2-wk reduction of ambulatory activity attenuates peripheral insulin sensitivity. *J Appl Physiol* (1985). 2010;108(5):1034–40.
26. Breen L, Stokes KA, Churchward-Venne TA, et al. Two weeks of reduced activity decreases leg lean mass and induces “anabolic resistance” of myofibrillar protein synthesis in healthy elderly. *J Clin Endocrinol Metabol*. 2013;98(6):2604–12.
27. Paton CM, Brandauer J, Weiss EP, et al. Hemostatic response to postprandial lipemia before and after exercise training. *J Appl Physiol*. 2006;101(1):316–21.
28. Hardman AE, Lawrence JE, Herd SL. Postprandial lipemia in endurance-trained people during a short interruption to training. *J Appl Physiol* (1985). 1998;84(6):1895–901.
29. Tudor-Locke C, Johnson WD, Katzmarzyk PT. Accelerometer-determined steps per day in US adults. *Med Sci Sports Exerc*. 2009;41(7):1384–91.
30. Costill DL. Metabolic responses during distance running. *J Appl Physiol*. 1970;28(3):251–5.
31. Tudor-Locke C, Bassett DR Jr. How many steps/day are enough? Preliminary pedometer indices for public health. *Sports Med*. 2004;34(1):1–8.
32. Frayn K. Calculation of substrate oxidation rates in vivo from gaseous exchange. *J Appl Physiol*. 1983;55(2):628–34.
33. Fielding BA, Callow J, Owen RM, Samra JS, Matthews DR, Frayn KN. Postprandial lipemia: the origin of an early peak studied by specific dietary fatty acid intake during sequential meals. *Am J Clin Nutr*. 1996;63(1):36–41.
34. McGuire S. US Department of Agriculture and US Department of Health and Human Services, Dietary Guidelines for Americans, 2010. Washington (DC): U.S. Government Printing Office, January 2011. *Adv Nutr*. 2011;2(3):293–4.
35. Freese EC, Gist NH, Cureton KJ. Effect of prior exercise on postprandial lipemia: an updated quantitative review. *J Appl Physiol*. 2013;116(1):67–75.
36. Duvivier BM, Schaper NC, Bremers MA, et al. Minimal intensity physical activity (standing and walking) of longer duration improves insulin action and plasma lipids more than shorter periods of moderate to vigorous exercise (cycling) in sedentary subjects when energy expenditure is comparable. *PLoS One*. 2013;8(2):e55542.
37. Ekelund U, Steene-Johannessen J, Brown WJ, et al. Does physical activity attenuate, or even eliminate, the detrimental association of sitting time with mortality? A harmonised meta-analysis of data from more than 1 million men and women. *Lancet*. 2016;388(10051):1302–10.
38. Shad BJ, Thompson JL, Holwerda AM, et al. One week of step reduction lowers myofibrillar protein synthesis rates in young men. *Med Sci Sports Exerc*. 2019;51(10):2125–34.
39. Wolfe AS, Burton HM, Vardarli E, Coyle EF. Hourly 4-s sprints prevent impairment of postprandial fat metabolism from inactivity. *Med Sci Sports Exerc*. 2020;52(10):2262–9.
40. Tudor-Locke C, Craig CL, Thyfault JP, Spence JC. A step-defined sedentary lifestyle index: <5000 steps/day. *Appl Physiol Nutr Metab*. 2013;38(2):100–14.
41. Kraus WE, Janz KF, Powell KE, et al. Daily step counts for measuring physical activity exposure and its relation to health. *Med Sci Sports Exerc*. 2019;51(6):1206–12.
42. Lee IM, Shiroma EJ, Kamada M, Bassett DR, Matthews CE, Buring JE. Association of step volume and intensity with all-cause mortality in older women. *JAMA Intern Med*. 2019;179(8):1105–12.
43. Yates T, Henson J, Khunti K, et al. Effect of physical activity measurement type on the association between walking activity and glucose regulation in a high-risk population recruited from primary care. *Int J Epidemiol*. 2013;42(2):533–40.
44. Hamilton MT, Hamilton DG, Zderic TW. Role of low energy expenditure and sitting in obesity, metabolic syndrome, type 2 diabetes, and cardiovascular disease. *Diabetes*. 2007;56(11):2655–67.
45. Church TS, Thomas DM, Tudor-Locke C, et al. Trends over 5 decades in U.S. occupation-related physical activity and their associations with obesity. *PLoS One*. 2011;6(5):e19657.
46. Whitfield G, Pettee Gabriel KK, Kohl HW. Sedentary and active: self-reported sitting time among marathon and half-marathon participants. *J Phys Act Health*. 2014;11(1):165–72.
47. Jeffers BJ, Sartini C, Shiroma E, Whincup PH, Wannamethee SG, Lee IM. Duration and breaks in sedentary behaviour: accelerometer data from 1566 community-dwelling older men (British Regional Heart Study). *Br J Sports Med*. 2015;49(24):1591–4.
48. Wang H, Eckel RH. Lipoprotein lipase: from gene to obesity. *Am J Physiol Endocrinol Metab*. 2009;297(2):E271–88.
49. Bey L, Hamilton MT. Suppression of skeletal muscle lipoprotein lipase activity during physical inactivity: a molecular reason to maintain daily low-intensity activity. *J Physiol*. 2003;551(Pt 2):673–82.
50. Kawamoto E, Tamakoshi K, Ra S-G, Masuda H, Kawanaka K. Immobilization rapidly induces thioredoxin-interacting protein gene expression together with insulin resistance in rat skeletal muscle. *J Appl Physiol*. 2018;125(2):596–604.
51. Beigneux AP. GPIHBP1 and the processing of triglyceride-rich lipoproteins. *Clin Lipidol*. 2010;5(4):575–82.