

Postprandial lipoprotein profile in two modes of high-intensity intermittent exercise

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The aim of present study was to compare blood lipid postprandial profile response in two modes of high-intensity intermittent exercise. Twelve individuals (6 men and 6 women) were submitted to a maximal incremental test (to determine maximal aerobic power [MAP] and $\dot{V}O_{2peak}$ [peak oxygen uptake]), high-intensity intermittent all-out exercise (60×8-sec bouts interspersed by 12-sec passive recovery) and fixed high-intensity intermittent exercise (100% maximal aerobic speed, consisted of 1-min repetitions at MAP [70 rpm] separated by 1-min of passive recovery). Blood samples were collected pre, immediately, 45 and 90-min postexercise. Serum was analyzed for total cholesterol and its ratio, high-density lipoprotein cholesterol (HDL-c), low-density lipoprotein cholesterol (LDL-c), very low-density lipoprotein (VLDL) cholesterol,

and triacylglycerol (TAG). For TAG there was a main effect of moment with higher values immediately postexercise compared to 45-min postexercise. For VLDL there was a main effect to moment with higher values immediately post exercise than pre and 45-min postexercise; higher values 90-min postexercise than 45-min postexercise. There was no effect for HDL-c, LDL-c, and cholesterol. For area under the curve there was no difference for any variable. Our results indicated that both kinds of acute exercise session lead to no improvement in the acute response of serum lipid profile of healthy young.

Keywords: Exercise, Lipid metabolism, Physiology, Lipoproteins

INTRODUCTION

Physical inactivity is currently recognized as an epidemic since it affects a large part of the world population, from children to seniors (Bauman et al., 2012). This outcome is also related to the increased risk for several diseases, including insulin resistance, the leading cause of diabetes mellitus type 2, and dyslipidemia (Van Gaal et al., 2006).

Increased level of triacylglycerol is related to several chronic disorders, as well as all-cause mortality (Nordestgaard and Varbo, 2014). Indeed, triacylglycerol have been associated to 13% and 12% more odds of cardiovascular diseases and all-cause mortality, respectively (Liu et al., 2013). In addition, cardiovascular disease has a significant public health cost (Azambuja et al., 2008; de Oliveira et al., 2012).

In contrast, high-density lipoprotein cholesterol (HDL-c) is well established as a protector factor against cardiovascular disease (Rader and Hovingh, 2014). A classical study showed that an increase of 1 mg/dL in HDL-c was related to a significant decrease of coronary heart disease risk of more than 2% and a reduction of more than 3% of cardiovascular disease mortality rates, regardless of sex (Gordon et al., 1989). Therefore, interventions that improve lipoprotein profile are interesting regarding quality of life and public health concern.

A healthy lifestyle, especially regular exercise practice, protects against these chronic disorders, and is considered as a nonpharmacological treatment (Gupta et al., 1993; Lira et al., 2010; Lira et al., 2012; Tambalis et al., 2009). Recently, low-volume, high-in-

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tensity exercise has drawn attention, since it can result similar physiological and morphological effects, such as improvement of anaerobic and aerobic fitness, and body composition that those adaptations from moderate intensity exercise (Gibala, 2009; Gibala et al., 2009; Gibala et al., 2012; Gist et al., 2014; Hazell et al., 2014; Little et al., 2010). Moreover, a single session of high-intensity exercise was able to reduce total cholesterol and its fraction LDL-c (Lira et al., 2009).

Investigations have demonstrated benefits of high-intensity exercise performed in all-out manner (Gibala, 2009; Gibala et al., 2012), but this kind of exercise have particularities that make difficult its practical execution given that is necessary a specialized equipment as a mechanical cycle ergometer, treadmill or to be performed outside. Moreover, according to Gillen and Gibala (2014) such form of exercise may not be safe, tolerable or practical for many individuals. However, an alternative protocol, 1-min cycling efforts interspersed with 1 min of recovery, has showed positive results by fixed exercise intensity based in maximum indexes (i.e., maximum oxygen uptake or maximum load attained in incremental test) (Helgerud et al., 2007; Little et al., 2010), and also is feasible for healthy, overweight/obese and individuals with higher risk for cardiometabolic disorders (Gillen et al., 2013; Hood et al., 2011).

Indeed, the most efficient protocol to confer benefits is not known yet. The knowledge of the protocol that is able to maximize the acute benefits generated by exercise in lipid profile is important because this acute response can result in improve a long-term.

Therefore, the aim of the present study was analyze the effects of two high-intensity intermittent exercise cycling (HIIE), performed in all-out (HIIE-AO), and fixed intensity (HIIE-F) efforts upon lipoprotein profile in young men and women.

MATERIALS AND METHODS

Subjects

Twelve physically active men ($n=6$) and women subjects ($n=6$) (28 ± 2 years; 173 ± 12 cm; 73 ± 12 kg; peak oxygen uptake [$\dot{V}O_{2peak}$], 2.9 ± 0.8 L/min; maximal aerobic power [MAP]: 245 ± 71 W) participated in this study. Specific physical activities of the participants included strength training, recreational sports, cycling or running (2–3 times per week). Participants were included if they did not report any health problems and/or neuromuscular disorders that could affect their ability to complete the study protocol. Furthermore, all were free of any drug or ingestion of nutritional supplements during the period of the study and had

stable body mass 6 months before study ($\pm 5\%$ of body mass).

Participants took part voluntarily in the study after being informed about the procedures, risks, and benefits and signed an informed consent form. This study was approved by the local ethics committee (2012/133.609). The women were tested in the follicular phase (1–10 days after the onset of menstruation) of the menstrual cycle.

Study design

Subjects completed four experimental sessions separated by at least 72 hr. During the first session, anthropometric and $\dot{V}O_{2peak}$ test measurements on a cycle ergometer were taken. In the second session, the participants were submitted to a high-intensity, intermittent, all-out exercise protocol (60 bout of 8 sec: 12 sec) to determine the total work performed (kJ). As all-out exercise performance is difficult to predict and in order to guarantee that the results obtained were related to the type of exercise performed and not affected by the differences in exercise volume, total work performed was equalized based in total work done in this session. This value (kJ) was used to equalize the subsequent exercise sessions. The two experimental sessions remaining were applied in randomized order: a high-intensity intermittent exercise performed all-out and a high-intensity intermittent exercise with fixed load.

Anthropometric measurements

Height was measured using a stadiometer with a metric scale affixed to the floor, and body mass were measured using an electronic body-weight scale (precision 0.01 kg).

Maximum oxygen uptake test

The participants performed an incremental test to volitional exhaustion on a cycle ergometer (Lode B.V., Groningen, The Netherlands). The initial load was set at 30 W and it was increased by 25 W/min for men and 15 W/min for women. Cadence was set at 70 rpm, and subjects were instructed to perform the test until they could no longer continue. The test was finished when subjects could not maintain the load for 5 sec in the fixed cadence. Strong verbal encouragement was given during the test. The oxygen uptake was measured (MetaMax 3B, Cortex, Leipzig, Germany) throughout the test and the average of the last 30 sec was defined as $\dot{V}O_{2peak}$. The maximal load reached in the test was defined as the maximal intensity attained (MAP). When the subject was not able to finish the 1-min stage, the power was expressed according to the permanence time in the last stage, determined as the following: $MAP = \text{power of last stage completed} + [(time, in$

seconds, remaining in the last stage multiplied by 25 W or 15 W)/(60 sec].

Experimental protocol

Each experimental protocol started upon the arrival of the participant at approximately 8 a.m., and with the participant having fasted for at least 10 hr previously. A cannula was inserted in an antecubital vein, then a fasting blood sample was taken (time = zero). After this, participants received a standardized breakfast (25% of the estimated daily energy needs for each participant on a sedentary day (Mifflin et al., 1990)). This meal was composed of cheese, toast and strawberry yogurt and serial blood samples were taken at regular intervals for a period of 4 hr (2, 2.5, 3.25, and 4 hr). The exercise was performed at approximately 2–2.5 hr of the experiment.

Exercise protocols

In all exercise sessions participants performed a warm-up at 50% MAP for 5 min, and after a 2-min interval they started the exercise. To estimate the energy expenditure of all exercises the sum of the contribution of the three energy systems (phosphagen, glycolytic, and oxidative energy systems) was used (di Prampero and Ferretti, 1999). Estimates of oxidative, glycolytic, and phosphagen systems use were carried out through the measurement of oxygen consumption during activity, peak blood lactate concentration (measured from the ear lobe were taken to determine the lactate concentration - Yellow Spring 1500 Sport, Yellow Springs, United States) and the fast phase of excess oxygen consumption after exercise, respectively. The caloric quotient of 5.00 kcal was used in all three different energy systems.

Two high-intensity intermittent exercises were performed, an all out (HIIE-AO) and with fixed intensity (HIIE-F), equalized by the total work done that totalized 26.1 ± 9.8 kcal. The HIIE-F consisted of 1-min repetitions at MAP (70 rpm) separated by 1 min of passive recovery on a cycle ergometer until the completion of the previously determined exercise. The time spent on this exercise protocol was 17 ± 2 min (9 min of effort and 8 min of pause), and energy expenditure 161.9 ± 57.5 kcal. The HIIE-AO consisted in to cycle as fast as possible 60 times for 8 sec interspersed by 12 sec of passive recovery, totaling 20 min (8 min of effort and 12 min of pause), and energy expenditure 201.8 ± 64.1 kcal. The load used was 4% (men) and 2.5% (women) of body mass.

Blood samples and analysis

Blood samples were collected pre, immediately, 45- and 90-

min postexercise. The blood samples (10 mL) were immediately allocated into two 5-mL vacutainer tubes (Becton Dickinson, Juiz de Fora, MG, Brazil) containing ethylenediamine tetraacetic acid for plasma separation and into one 6-mL dry vacutainer tube for serum separation. The tubes were centrifuged at 3,500 rpm for 15 min at 4°C, and plasma and serum samples were stored at -20°C until analysis. Total cholesterol, HDL-c (mg/dL), and triacylglycerol (mg/dL) were assessed using commercial kits (Labtest, São Paulo, Brazil). LDL-c (mg/dL) and very low-density lipoprotein (VLDL) cholesterol (mg/dL) was calculated according to Friedewald et al. (1972). To eliminate inter-assay variance, all samples were analyzed in identical runs.

Statistical analysis

The data were presented as means and standard deviation. Linear mixed models were used to compare the lipid profile in different exercises across time (condition \times time). The Tukey *post hoc* was conducted if a significant difference was found. A paired *t*-test was conducted to compare area under curve (calculate by a trapezoidal method) between exercises. Statistical significance was set at $P < 0.05$. The data was analyzed using SAS 9.2 (SAS Institute Inc., Cary, NC, USA). Standardized effect sizes were also calculated by the Cohen equations (Cohen, 1988) with the following threshold values: < 0.2 , trivial; > 0.2 and < 0.6 , small; > 0.6 and < 1.2 , moderate; > 1.2 and < 2.0 , large; > 2.0 and < 4.0 , very large; < 4.0 , nearly perfect (Hopkins, 2002).

RESULTS

Fig. 1 presents blood levels of lipoprotein in the two modes of high-intensity intermittent exercise.

For postprandial triglyceride (TG) there was a tendency to main effect to condition ($F [1, 76] = 3.58, P = 0.062$) with higher values in HIIE-AO than HIIE-F ($P = 0.062, d = 0.416$ [small]). There was main effect of moment ($F [3, 76] = 5.18, P = 0.002$) with higher values immediately postexercise compared to 45-min postexercise ($P < 0.001, d = 0.307$ [small]). For VLDL there was a main effect of moment ($F [3, 69] = 3.08, P = 0.033$) with higher values immediately postexercise than pre ($P = 0.047, d = 0.211$ [small]), and 45-min postexercise ($P = 0.020, d = 0.281$ [small]); higher values 90-min postexercise than 45-min postexercise ($P = 0.027, d = 0.164$ [trivial]) and tendency to difference from pre exercise ($P = 0.063, d = 0.066$ [trivial]). There was no effect for HDL-c, LDL-c, and cholesterol. For area under the curve there was no difference for any variable.

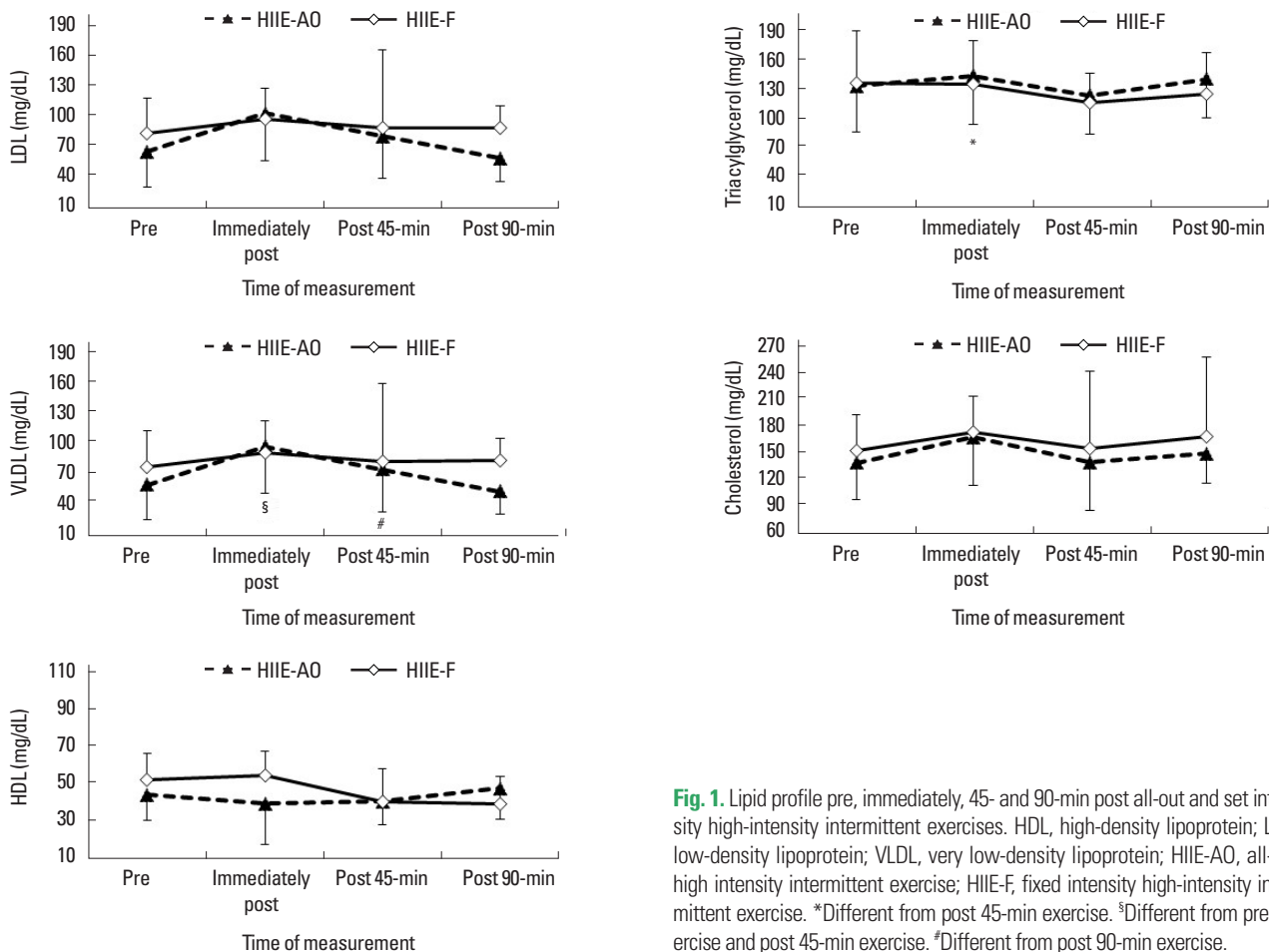


Fig. 1. Lipid profile pre, immediately, 45- and 90-min post all-out and set intensity high-intensity intermittent exercises. HDL, high-density lipoprotein; LDL, low-density lipoprotein; VLDL, very low-density lipoprotein; HIIE-AO, all-out high intensity intermittent exercise; HIIE-F, fixed intensity high-intensity intermittent exercise. *Different from post 45-min exercise. §Different from pre-exercise and post 45-min exercise. #Different from post 90-min exercise.

DISCUSSION

The main findings of the present study were that HIIE-F exercise seems to decrease TG when compared with HIIE-AO, and both exercises protocols induced similar responses for VLDL (increase postexercise). However, no other statistical significant improvement of lipoprotein profile was found in both exercise protocols.

These findings are in agreement with a recent study, which showed that a session composed by 20 all-out sprint lasting 6 sec of effort interspersed with 24-sec recovery did not change postprandial triacylglycerol, glucose and insulin in healthy men (Allen et al., 2014). The authors suggested that the lack of improvement for postprandial triacylglycerol was due the low energy expenditure of the exercise session (120 kcal approximately). In line with this results, the two modes of HIIE of the present study showed slight higher energy expenditure (162 to 202 kcal approximately) than the previous study (Allen et al., 2014), which may be one explanation for the lack of positive results in lipid profile.

However, a study showed that to achieve the positive effects of HIIE upon lipoprotein profile, especially in VLDL-TG secretion, the exercise should be performed for, at least, 2 months (Tsekouras et al., 2008), showing the importance of regular exercise in these outcomes. In addition, Lira et al. (2009) showed that high-intensity exercise (90% $\dot{V}O_{2max}$) performed continuously, with low energy expenditure (~124 kcal), decreased total cholesterol and LDL-c levels in young males.

Recently, Bellou et al. (2013) reported that HIIE (4 times of 4 min at 90% of $\dot{V}O_{2peak}$ interspersed with 4 min at 60% of $\dot{V}O_{2peak}$ for a total of 32 min; gross energy expenditure ~500 kcal) reduced fasting plasma VLDL concentrations in nonobese men the next day (14 hr after exercise bout) by augmenting VLDL clearance, just like a single bout of continuous endurance exercise. Therefore, further studies are clearly needed to identify the mechanisms underlying the beneficial effects of HIIE to help in the designing of optimal exercise protocols.

Alongside, energy expenditure seems to be a determinant in the

modulation of HDL concentrations after an exercise bout. It was suggested that to improve HDL the energy expenditure of the session should be higher than 1,100 kcal (Lira et al., 2012), although other study showed significant results upon HDL concentrations in acute exercise that resulted in more than 350 kcal (Park and Ransone, 2003). As previously discussed the energy expenditure in two HIIE protocols was not able to induce changes in HDL concentrations. However, it should be emphasized that the HDL concentration is a measurement of the total cholesterol content alone, carried in plasma HDL. Moreover, there is a heterogeneity in the HDL composition (Fruchart et al., 1993), as well as, there are HDL subfractions that are more likely to suffer the effects of acute exercise (Campbell et al., 2011; Frey et al., 1993; Park and Ransone, 2003). As we did not evaluate such outcome, this is a limitation of the present study.

Another possible explanation to the no improvement in lipid profile may be the absence of chronic disorders, such as obesity, dyslipidemia and insulin resistance in the participants of the present study. Freese et al. (2015) showed that four all-out bouts (Wingate test), performed acutely prior and after 6-week training, resulted in no changes in fasting triacylglycerol in middle-aged women with metabolic syndrome. However, when postprandial lipemia was analyzed all-out bouts resulted in 13.1% decrease in this group. Such outcome probably occurred because women with metabolic syndrome present higher postprandial lipemia than their healthy peer (Kolovou et al., 2006), and thus, be more likely to show changes.

Chronic exercise has widely been demonstrated to reduce risk of metabolic and cardiovascular disorders associated with cardiovascular diseases (Lira et al., 2012; Tambalis et al., 2009). Furthermore, there are sustainable evidences showing that aerobic exercise is one of the most effective in improving lipoprotein profile regarding the population (Tambalis et al., 2009). However, since obesity rates remain increasing in several countries and the free-living time for exercise practice is diminishing, the HIIE protocols emerge as a low financial cost and time-efficient exercise and way to help in the prevention and probably treatment for many chronic diseases, especially by its ability to improve physical fitness and body composition. However, the most efficient HIIE protocol to improve lipid profile remains yet not established.

Finally, it is possible that due the dependence of glucose as a substrate to maintain exercise in high intensity, the mechanisms involved in this type of acute exercise do not seem to be the predominant modulator of the lipid profile (van Loon et al., 2001). In summary, our results indicate that both kinds of acute exercise

session lead to no improvement in serum lipid profile of healthy young.

CONFLICT OF INTEREST

No potential conflict of interest relevant to this article was reported.

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