Accepted Manuscript

Accumulating exercise and postprandial health in adolescents

Bert Bond, Craig A. Williams, Sarah R. Jackman, Adam Woodward, Neil Armstrong, Alan R. Barker

PII: S0026-0495(15)00160-2
DOI: doi: 10.1016/j.metabol.2015.05.016
Reference: YMETA 53219

To appear in: Metabolism

Received date: 6 February 2015
Revised date: 5 May 2015
Accepted date: 8 May 2015

Please cite this article as: Bond Bert, Williams Craig A., Jackman Sarah R., Woodward Adam, Armstrong Neil, Barker Alan R., Accumulating exercise and postprandial health in adolescents, Metabolism (2015), doi: 10.1016/j.metabol.2015.05.016

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.
Accumulating exercise and postprandial health in adolescents

Bert Bond¹, Craig A. Williams¹, Sarah R. Jackman², Adam Woodward¹, Neil Armstrong¹ and Alan R. Barker¹

¹Children’s Health and Exercise Research Centre, Sport and Health Science, College of Life and Environmental Sciences, University of Exeter, Exeter, EX1 2LU.

²Sport and Health Science, College of Life and Environmental Sciences, University of Exeter, Exeter, EX1 2LU.

Corresponding author:

Dr Alan R. Barker

Children’s Health and Exercise Research Centre, Sport and Health Sciences
College of Life and Environmental Sciences
University of Exeter
St Luke’s Campus
Exeter
EX1 2LU

Tel: 44 (0)1392 722766
Fax: 44 (0)1392 724726
Email: A.R.Barker@exeter.ac.uk

Word count of abstract: 249

Number of references: 39

Number of tables and figures: 7

This study was supported by the Sport and Health Sciences Research Committee, College of Life and Environmental Sciences, University of Exeter.

The authors confirm the absence of any conflicts of interest.
ABSTRACT

**Purpose:** To examine the influence of exercise intensity on postprandial health outcomes in adolescents when exercise is accumulated throughout the day. **Methods:** 19 adolescents (9 male, 13.7 ± 0.4 y) completed three 1-day trials in a randomised order: 1) rest (CON); or four bouts of 2) 2 x 1 min cycling at 90% peak power with 75 s recovery (high-intensity interval exercise; HIIE); or 3) cycling at 90% of the gas exchange threshold (moderate-intensity exercise; MIE), which was work-matched to HIIE. Each bout was separated by 2 hours. Participants consumed a high fat milkshake for breakfast and lunch. Postprandial triacylglycerol (TAG), glucose, systolic blood pressure (SBP) and fat oxidation were assessed throughout the day. **Results:** There was no effect of trial on total area under the curve (TAUC) for TAG (\(P=0.87\)). TAUC-glucose was lower in HIIE compared to CON (\(P=0.03, ES=0.42\)) and MIE (\(P=0.04, ES=0.41\)), with no difference between MIE and CON (\(P=0.89, ES=0.04\)). Postprandial SBP was lower in HIIE compared to CON (\(P=0.04, ES=0.50\)) and MIE (\(P=0.04, ES=0.40\)), but not different between MIE and CON (\(P=0.52, ES=0.11\)). Resting fat oxidation was increased in HIIE compared to CON (\(P=0.01, ES=0.74\)) and MIE (\(P=0.05, ES=0.51\)), with no difference between MIE and CON (\(P=0.37, ES=0.24\)). **Conclusion:** Neither exercise trial attenuated postprandial lipaemia. However, accumulating brief bouts of HIIE, but not MIE, reduced postprandial plasma glucose and SBP, and increased resting fat oxidation in adolescent boys and girls. The intensity of accumulated exercise may therefore have important implications for health outcomes in youth.

**Key words**
Cardiovascular disease (CVD), exercise intensity, young people, physical activity.
### ABREVIATIONS

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>CVD</td>
<td>Cardiovascular disease</td>
</tr>
<tr>
<td>EE</td>
<td>Energy expenditure</td>
</tr>
<tr>
<td>ES</td>
<td>Effect size</td>
</tr>
<tr>
<td>GET</td>
<td>Gas exchange threshold</td>
</tr>
<tr>
<td>HFM</td>
<td>High fat meal</td>
</tr>
<tr>
<td>HIIE</td>
<td>High-intensity interval exercise</td>
</tr>
<tr>
<td>IAUC</td>
<td>Incremental area under the curve</td>
</tr>
<tr>
<td>MIE</td>
<td>Moderate-intensity exercise</td>
</tr>
<tr>
<td>PACES</td>
<td>Physical activity enjoyment scale</td>
</tr>
<tr>
<td>PPL</td>
<td>Postprandial lipaemia</td>
</tr>
<tr>
<td>REE</td>
<td>Resting energy expenditure</td>
</tr>
<tr>
<td>SBP</td>
<td>Systolic blood pressure</td>
</tr>
<tr>
<td>TAG</td>
<td>Triacylglycerol</td>
</tr>
<tr>
<td>$\hat{V}O_2$ max</td>
<td>Maximal oxygen uptake</td>
</tr>
</tbody>
</table>
1. INTRODUCTION

Repeat exposure to elevated postprandial plasma triacylglycerol (TAG) and glucose concentrations has been implicated in the pathogenesis of type two diabetes [1] and atherosclerosis [2], which have their origins in youth [3, 4]. Elevated non-fasting plasma TAG, glucose, and systolic blood pressure (SBP) in adolescence are independently associated with fatty streaks in the coronary arteries and future cardiovascular risk [5-7]. Furthermore, postprandial hypertension has been purported as a novel risk factor for atherosclerosis in adults [8]. Considering that most of the day may be spent in the postprandial state, it is important to identify feasible interventions to modulate these risks factors for cardiovascular disease in youth.

We have recently demonstrated that a single bout of exercise (~ 30 min) can improve postprandial health outcomes in adolescents in an intensity-dependent manner [9]. However, it is known that adolescents rarely sustain exercise for longer than 10 min [10]. Therefore it is important to address whether accumulating short bouts of exercise over the course of the day can favourably modulate postprandial health in this group.

It has been demonstrated that performing brief (3-10 min) bouts of low to moderate intensity exercise throughout the day may reduce postprandial plasma TAG to the same extent [11], or greater than [12], an equivalent volume of continuous exercise in adults. Similar exercise patterns have also been shown to lower SBP in normotensive adults [11]. Accumulating moderate-intensity exercise (MIE) the day before a high fat meal has been shown to lower postprandial plasma glucose and TAG concentrations, and improve endothelial function in adolescent boys [13]. However, the timing of the exercise stimulus in relation to a high fat meal is known to effect the subsequent lipaemia [14], and no study with adolescents has addressed the impact of exercise accumulated on the same day as the test meal, or if the
intensity of accumulated exercise influences the postprandial response. The latter point is important to consider as there is evidence showing that performing high-intensity exercise is superior than MIE at modifying cardiometabolic risk factors in youth [15, 16], even when the total amount of high-intensity exercise performed is small (~ 4 min) [17].

Given the above, the purpose of this investigation was to test the hypothesis that accumulating short bouts of work matched high-intensity interval exercise (HIIE) and MIE would improve parameters of postprandial health in youth (e.g. plasma glucose and TAG, blood pressure and lipid oxidation), but the benefits would be superior in HIIE compared to MIE. The present study also builds on our earlier work by identifying whether comparable benefits are achievable when the same exercise stimulus is accumulated in smaller bouts rather than performed in a single session [9].

2. METHODS

2.1 Participants: Twenty one 13 to 14-year-old adolescents (11 boys) initially volunteered to take part in this study. Participant assent and parental consent were provided before participation in the project, which was approved by the institutional ethics committee (reference number 2012/391). Exclusion criteria included any contraindications to exercise, the presence of disease or musculoskeletal injury and the use of any medication or substance known to influence carbohydrate or fat metabolism. These criteria precluded two boys from taking part (diagnosed asthma n=1; use of medication n=1), thus only 19 adolescents (9 boys) completed this investigation.

2.2 Experimental protocol: This study required four visits to the laboratory over a period of three weeks, each separated by at least 4 days, and incorporated a within measures design. All
exercise tests were performed using an electronically braked cycle ergometer (Lode Excallibur Sport, Groningen, the Netherlands).

**Visit 1: Maximal oxygen uptake (\( \dot{V}O_2 \text{max} \)) and gas exchange threshold (GET) determination**

Body mass and stature were measured to the nearest 0.1 kg and 0.01 m respectively. Body mass index (BMI) was interpreted using established cut points for this population [18]. Percentage body fat was estimated using triceps and subscapular skinfold thickness according to Slaughter *et al.* [19] and pubertal status was determined by a self-assessment of secondary sexual characteristics of pubic hair development [20].

Participants were habituated to exercise on the cycle ergometer before completing a validated combined ramp and supramaximal test to exhaustion to establish \( \dot{V}O_2 \text{max} \) [21]. Pulmonary \( \dot{V} O_2 \) was monitored throughout (Coretex Metalyzer III B, Leipzig, Germany) and the GET was identified as the disproportionate increase in carbon dioxide production (\( \dot{V}CO_2 \)) relative to \( \dot{V} O_2 \) and an increase in expired ventilation (\( \dot{V}E/\dot{V}O_2 \)) with no increase in \( \dot{V}E/\dot{V}CO_2 \). \( \dot{V}O_2 \text{max} \) was determined as the highest 10 second average in \( \dot{V} O_2 \) elicited either during the ramp test or supramaximal bout. Aerobic fitness was interpreted using current thresholds for metabolic health [22].

**Visits 2-4: Experimental trials**

A schematic of each trial is provided in Figure 1. Following a ~12 h overnight fast, participants arrived at the laboratory at 07:45 and rested for 10 min before providing a fasting capillary blood sample for plasma glucose and TAG concentrations. At 08:00 SBP was recorded after spending ~ 10 min in a seated position using an automated inflation cuff (Dinamap CareScap V100, GE Healthcare, USA). Resting metabolic rate (RMR) was then
assessed via indirect calorimetry (Cortex Metalyzer 3B, Leipzig, Germany) for 15 min in order to determine total resting energy expenditure (REE) and substrate oxidation (lipid and carbohydrate) in accordance with our earlier work [9, 23]. These measures were repeated 45 min after the cessation of each exercise bout or rest.

On three separate occasions, approximately one week apart, participants completed in a randomised order: 1) two, 1 minute intervals at 90% of the peak power determined from the ramp test, separated by 75 s at 20 W (HIIE); 2) ~ 6 min of cycling at 90% GET (MIE); or 3) remained seated and watched films in the laboratory (control; CON). The exercise bouts were repeated four times, each separated by two hours (see Figure 1). A warm up and cool down of 75 s at 20 W was included for each HIIE and MIE. The duration of the MIE trial was calculated to match the total work performed during the HIIE bout for each participant, and the total accrued exercise stimulus matched our earlier postprandial investigation with this group [9]. Total EE and the macronutrient energetic contributions to MIE were determined using the mean exercise $\dot{V}O_2$ and respiratory exchange ratio (RER) values [24]. Protein oxidation was assumed to be negligible, and an RER >1 was taken to represent 100% carbohydrate oxidation.

Participants were asked to provide a rating of perceived exertion (RPE) using the 1-10 Pictorial Children’s Effort Rating Table [25] in the final 10 s of exercise. Participants also completed the 16-point Physical Activity Enjoyment Scale (PACES) [26] and identified which exercise trial they preferred upon immediate completion of the final exercise bout.

**2.3 High fat meal (HFM) and postprandial observation:** Participants consumed a milkshake of three parts Cornish ice cream and one part double cream between 08:45 and
09:00. An identical milkshake was consumed between 12:45 and 13:00. The milkshake provided approximately 1.50 g of fat (70% total energy), 1.20 g carbohydrate (25%) and 0.21 g of protein (5%) per kilogram of body mass (80 kJ·kg BM\(^{-1}\)) in accordance with our prior investigation with adolescents [9]. No other food was consumed during the postprandial period, although water was available ad libitum and subsequently replicated for each trial.

### 2.4 Blood analyses:
For each blood collection, ~ 600 µL of capillary blood was collected into lithium-heparin coated (TAG) and heparin-fluoride coated (glucose) Microvette CB 300 tubes (Sarstedt Ltd, Leicester, UK) and centrifuged immediately at 13,000 g for 15 min. Plasma was then removed and either stored at -80°C for one month for TAG analysis, or analysed immediately using a YSI 2300 Stat Plus Glucose and L-Lactate Analyzer (YSI Inc., Yellow Springs, USA) for glucose. Plasma TAG was quantified in duplicate by enzymatic, colorimetric methods using an assay kit according to the manufacturer’s guidelines (Cayman Chemical Company, MI, USA). The within-batch coefficients of variation for plasma TAG and glucose were 1.8% and 0.6% respectively.

### 2.5 Standardisation of physical activity and diet:
With parental supervision, participants were asked to wear an ActiGraph GT1M accelerometer (ActiGraph, LLC, Pensacola, USA) and complete a food diary during the 48 h period immediately preceding each laboratory visit. Participants were asked to replicate their diet prior to each laboratory visit and were verbally reminded of this requirement. The food diaries were subsequently assessed for total energy and macronutrient intakes (CompEat Pro, Nutrition Systems, UK). Time spent performing moderate to vigorous activity was determined using established cut points for paediatric groups [27].
2.6 Statistical analyses: The total area under the curve (TAUC) analysis was performed using the trapezium rule (GraphPad Prism, GraphPad Software, San Diego, CA) to describe the changes in plasma TAG, glucose, SBP, REE and fat oxidation. The incremental area under the curve (IAUC) was also calculated for plasma TAG and glucose in order to characterise the magnitude of the response and the changes over time. All plasma TAG and glucose area under the curve analyses were calculated using the time point immediately before the first HFM.

Descriptive statistics were calculated using SPSS (version 19.0, Chicago, USA) and presented as mean ± SD. Mean differences in descriptive statistics between boys and girls were analysed using independent samples t tests. The mean differences in the physiological and perceptual responses of the boys and girls during HIIE and MIE were analysed using paired samples t tests. Analysis of fasting plasma TAG and glucose, and AUC analyses for plasma TAG, plasma glucose, fat oxidation and SBP were performed using a mixed model ANOVA with trial (CON, MIE, HIIE) and sex (male, female) as the main effects. A main effect for sex was only present when analysis plasma TAG, meaning all other variables are reported with boys and girls combined into a single ANOVA model. Normality of distribution was checked using the Shapiro-Wilk test, and data were log transformed if this assumption was violated. Homogeneity of variance was determined using Mauchly’s test of sphericity and the degrees of freedom were adjusted using the Greenhouse-Geisser correction if required. Pairwise comparisons between means were interpreted using the P value, 95% confidence intervals (CI) and standardised effect sizes (ES). The null hypothesis was rejected at an alpha level of 0.05, and an ES of 0.20, 0.50 and 0.80 was considered to represent a small, moderate and large change between means [28]. Relationships between changes in AUC outcomes for TAG and potentially mechanistically important variables (e.g.
postprandial resting fat oxidation) were explored using Pearson’s correlation coefficients and their associated $P$ value.

3. RESULTS

Baseline participant characteristics are presented in Table 1. The sexual maturation status for boys and girls was as follows: Tanner stage 3, $n=5$ and $n=4$; Tanner stage, 4 $n=3$ and $n=6$; Tanner stage 5, $n=1$ and $n=0$. Two boys and 2 girls were overweight, and 5 boys and 2 girls did not achieve the recommended aerobic fitness level for metabolic health. No differences in energy intake, individual macronutrient contributions, or time spent performing moderate to vigorous physical activity were apparent for boys or girls during the 48 h preceding each laboratory visit ($P>0.74$, $ES<0.20$; Table 2). Only 2 boys and 1 girl achieved the recommended minimum of $> 60$ minutes of moderate to vigorous physical activity per day.

3.1 Exercise protocols: Table 3 presents the physiological and perceptual data from the exercise trials. The average length of each MIE bout was $6.8 \pm 1.2$ min for boys and $5.5 \pm 1.2$ min for girls. The total accumulated exercise time in the MIE trial was $29.6 \pm 4.9$ min and $23.6 \pm 4.3$ min for boys and girls respectively. Eight boys but only 3 girls indicated that they preferred the HIIE exercise bout.

3.2 Plasma TAG: There were no differences in fasted plasma TAG concentrations between trials for boys or girls (main effect for trial, $P=0.74$; main effect for sex, $P=0.07$; trial by sex interaction, $P=0.33$). Mean plasma TAG concentrations during the postprandial period are illustrated in Figure 2 and the AUC analyses are described in Table 4. There was an effect of sex, but not trial, on TAUC-TAG (main effect for trial, $P=0.87$; main effect for sex, $P=0.01$;
trial by sex interaction, \( P=0.29 \), with mean TAUC-TAG lower for girls than boys across all trials \( (P=0.01, 95\% \text{ CI } -2.96 \text{ to } -0.50, ES=1.25) \). There was also an effect of sex, but not trial, on IAUC-TAG (main effect for trial, \( P=0.61 \); main effect for sex, \( P=0.01 \); trial by sex interaction, \( P=0.18 \)), with mean IAUC-TAG across all trials lower for girls than boys \( (P=0.01, 95\% \text{ CI } -2.39 \text{ to } -0.35, ES=1.21) \). There were no effects of trial on TAUC-TAG or IAUC-TAG during the 4 hours after the first \( (P=0.74 \text{ and } P=0.24) \) or second \( (P=0.95 \text{ and } P=0.17) \) HFM. Relationships between \( \dot{V}O_2 \text{ max, body fat, EE, fat oxidation or mean } \dot{V}O_2 \) and IAUC-TAG were not statistically significant for boys or girls \( (r < 0.2, P>0.16 \text{ for all}) \).

### 3.3 Plasma glucose

Mean plasma glucose concentrations are depicted in Figure 2 and the AUC analyses are provided in Table 4. Fasted plasma glucose was not different between trials \( (P=0.46) \). There was a strong trend for a main effect of trial on TAUC-glucose \( (P=0.05) \), and pairwise comparisons revealed that TAUC-glucose was lower in HIIE compared to CON \( (P=0.03, 95\% \text{ CI } -1.64 \text{ to } -0.08, ES=0.42) \) and MIE \( (P=0.04, 95\% \text{ CI } -1.57 \text{ to } -0.04, ES=0.41) \), with no difference between MIE and CON \( (P=0.89, 95\% \text{ CI } -0.93 \text{ to } 0.82, ES=0.04) \). There was no effect of trial on the 4 hour TAUC-glucose after the first HFM \( (P=0.16) \), but there was a difference after the second HFM \( (P=0.03) \) with TAUC-glucose lower in HIIE compared to CON \( (P=0.01, 95\% \text{ CI } -1.02 \text{ to } -0.15, ES=0.61) \) and MIE \( (P=0.02, 95\% \text{ CI } -0.89 \text{ to } -0.10, ES=0.55) \), but no difference between MIE and CON \( (P=0.75, 95\% \text{ CI } -0.67 \text{ to } 0.49, ES=0.09) \). There was no main effect of trial for IAUC-glucose \( (P=0.49) \), or for the IAUC-glucose during the 4 hours after the first \( (P=0.90) \) and second \( (P=0.55) \) HFM.

### 3.4 Postprandial blood pressure

There was a strong trend for a main effect for trial on postprandial TAUC-SBP \( (P=0.05; \text{ Figure 3}) \). TAUC-SBP was lower in HIIE compared to
CON ($P=0.02$, 95% CI -31 to -3, $ES=0.31$) but not different to MIE ($P=0.10$, 95% CI -28 to 3, $ES=0.23$). TAUC-SBP was not different between MIE and CON ($P=0.54$, 95% CI -18 to 10, $ES=0.09$).

3.5 REE and fat oxidation: There was no main effect of trial on TAUC-REE ($P=0.29$, data not presented). There was a main effect for trial on postprandial TAUC-Fat ($P=0.01$; Figure 3). TAUC-Fat oxidation increased in HIIE by 17% compared to CON ($P=0.01$, 95% CI 3 to 13, $ES=0.74$) and by 11% compared to MIE ($P=0.048$, 95% CI 0 to 11, $ES=0.51$). There was no difference in TAUC-Fat oxidation between MIE and CON ($P=0.37$, 95% CI -3 to 8, $ES=0.24$). Changes in TAUC-TAG and IAUC-TAG were not related to postprandial TAUC-fat oxidation (all $r < 0.2$).

4. DISCUSSION
This study is the first to isolate the effect of exercise intensity on postprandial health outcomes in adolescent girls and boys when exercise is accumulated on the same day as two high fat meals. The novel findings of this study are: 1) accumulating small volumes of HIIE and MIE throughout the day did not attenuate postprandial lipaemia; and 2) repeating brief HIIE, but not MIE, lowered postprandial plasma glucose concentrations and SBP, and increased resting fat oxidation in adolescent boys and girls. This study demonstrates that even small volumes (8 min) of exercise performed throughout the day may favourably modulate postprandial health outcomes such as plasma glucose, SBP and fat oxidation in a manner which is intensity-dependent.

We have previously shown that the same total MIE and HIIE stimulus performed in a single session 1 hour before an identical HFM can attenuate PPL in girls but not boys [9]. However,
we did not observe any reduction in postprandial plasma TAG in the present study. We are aware of only two studies which have investigated the efficacy of accumulating exercise on the same day as a series of high fat meals [29, 30], and these authors reported only a ~10% reduction in TAUC-TAG was achievable in adults when accruing a total of 30 minutes of exercise at 60-70% $\dot{V}O_2_{max}$. In agreement with our data, a recent study has shown that performing 135 min of walking during the postprandial period did not reduce plasma TAG concentrations in adolescent boys and girls [31]. Furthermore, Zhang et al. have previously demonstrated in adults that PPL was only attenuated when exercise was performed 1 hour before a HFM, compared to when identical exercise was performed during the postprandial period [14]. Therefore, accumulating exercise during the postprandial period may only have a limited effect on PPL. Interestingly, 75% of the exercise stimulus in the present study was completed before the consumption of the second HFM, however there were no differences between trials in the 4 hour TAUC-TAG after the first or the second HFM. Accumulating exercise has been shown to lower PPL the following day in adults [11, 12] and adolescents [13], although the latter did not reach statistical significance. Therefore it is likely that repeating brief bouts of exercise may have some utility in attenuating PPL several hours after the final exercise bout, but further work is needed to establish whether this effect can be modulated by exercise intensity.

An interesting finding of the present study is the reduction in TAUC-glucose in the HIIE trial, but not MIE, compared to CON. We acknowledge that the absolute differences in TAUC-glucose between CON and MIE was small ($ES=0.42$ and 0.41, respectively), and this probably reflects the high fat content of the test meal and that our participants were normoglycaemic. It is also not known how the magnitude of this reduction in TAUC-glucose affects cardiometabolic risk if repeated on a daily basis. However, this is the first study to identify that an independent effect of exercise intensity exists when exercise bouts are brief,
which may be important considering that young people rarely exercise for longer than 10 min [10]. Consequently, future studies exploring this interaction between intensity and accumulated exercise are warranted.

It is important to note that we have previously failed to report an effect of exercise on plasma glucose concentrations after a single bout of MIE or HIIE (equating to the total exercise stimulus accrued in the present study) performed one hour before an identical test meal [9]. Whilst we are unable to provide any mechanistic insight regarding why accumulating HIIE exercise may be more efficacious than performing the same total exercise stimulus in a single bout, studies with adults have shown that repeatedly interrupting sitting time can lower postprandial plasma glucose concentrations [32], and that minimising continuous sedentary behaviour is important for metabolic health independent of physical activity [33]. This suggests that the repetitive pattern of exercise in the present study may play an important role in promoting glucose control, although this does not explain the absence of an effect in MIE. Interestingly, we recently demonstrated that a similar bout of MIE delivered in a single session offered comparable reductions in TAUC-glucose as HIIE one hour before an oral glucose tolerance test [23]. Therefore, exercise intensity may be an important factor regarding glycaemic control when the duration of exercise is short. This finding is consistent with existing literature identifying that accumulating even small volumes (4-8 minutes) of high-intensity (but not moderate-intensity) exercise is associated with a reduction in cardiometabolic risk in adolescents [15, 17]. Given that adolescents often fail to achieve the recommended 60 minutes of physical activity [34], and the duration of exercise bouts performed by young people rarely last longer than 10 minutes [10], it would appear that HIIE has some utility in promoting glycaemic control in adolescents.
Postprandial hypertension is purported to be a novel risk factor for atherosclerosis in adults [8], and elevated SBP during adolescence is associated with future cardiovascular disease [7]. Our data indicate that reductions in postprandial SBP are achievable when only two minutes of HIIE, but not MIE, are performed on four occasions over the course of the day. Whilst we are unable to shed light on the mechanism(s) underlying this intensity-dependent response in postprandial SBP, it has been demonstrated that 2 x 15 min of high, but not moderate, intensity exergaming influences vascular function in children [35]. Therefore, changes in SBP in HIIE may be related to an improvement in endothelial function and/or a reduction in vascular resistance. Further work is needed to identify the acute influence of exercise intensity on vascular function in this group, but it has been shown that accumulating exercise is equally effective in lowering SBP the following day as an equivalent bout of continuous exercise in adults [11]. In contrast, the magnitude of the reduction in postprandial SBP observed in the accumulated HIIE trial in the present study is lower than what we have previously reported when HIIE is performed in a single bout (ES=0.31 vs 0.68) [9]. However, the present findings are promising considering the very brief nature of each HIIE bout, and our data adds to a growing body of literature indicating that low volumes of HIIE may be effectual in lowering SBP even in normotensive adolescents [9, 36, 37].

We have previously demonstrated that HIIE, but not MIE, increases postprandial fat oxidation during the immediate hours after exercise in adolescent boys and girls [9]. The findings of the current study indicate that this favourable shift in resting substrate utilisation may also occur when small volumes of HIIE, but not MIE, are accumulated throughout the day in adolescents. Additionally, the effect sizes between our earlier work and the present study are broadly comparable (ES=0.88 and 0.74, respectively). This finding may have clinical importance as repeating 2 minute bouts of HIIE is likely to be achievable for most
adolescents, and an elevation in resting fat oxidation is an important predictor of the magnitude of exercise-induced fat loss [38], and may be linked to insulin sensitivity [39].

It is of interest that the PACES score for HIIE and MIE were comparable for both boys and girls, despite the greater perceptual and physiological exertion in the HIIE. Our findings are arguably limited to a stringently controlled, laboratory-based protocol and therefore lack ecological validity, however preliminary findings of other laboratories [40, 41] and school-based [36] interventions suggest that HIIE may be a feasible alternative for health promotion in adolescent boys and girls.

This study is the first to demonstrate that accumulating exercise may provide small benefits in postprandial health in a manner which is intensity-dependent. Whilst we are unable to provide any mechanistic insight regarding the independent effect of exercise intensity, or translate these effects into clinical relevance, our findings are conceptually important as adolescents rarely perform exercise for longer than 10 minutes [10], and most of the day may be spent in the postprandial state. Thus, investigations which identify how this pattern of exercise can be optimized are warranted. Cross-sectional data are available indicating that achieving approximately 7 minutes of vigorous intensity physical activity per day (but not ~ 110 minutes of light or ~ 46 minutes of moderate intensity) reduces cardiometabolic risk in adolescents [15]. Furthermore, longitudinal evidence in young people indicates that performing ~ 4 minutes of vigorous activity per day may lower cardiovascular disease risk over time [17]. Therefore, it is plausible that the small effects observed in the HIIE trial may provide meaningful health benefits if performed on a regular basis.
The strengths of this study include the isolation of exercise intensity, the adoption of multiple meals and the comparison between boys and girls. Furthermore, the brief exercise bouts in this investigation more accurately reflect the pattern of physical activity performed by this age group than continuous exercise of a longer (>10 min) duration [10]. Therefore, identifying that the intensity of accumulated exercise intensity is important regarding postprandial health is likely to be an important health message. However, these data should be considered in light of a number of limitations. Firstly, the test meals adopted in this study bear little resemblance to a typical diet. It is therefore important to see if our findings can be replicated following the consumption of more representative meals in a larger sample size. Secondly, we cannot extrapolate our findings beyond our sample population. Further work is required to establish the influence of the intensity of accumulated exercise on these outcomes in adolescents with cardiometabolic risk factors. Finally, we were not able to assess the influence of the MIE and HIIE bouts on these parameters the following day. Given that exercise accumulated the day beforehand has been shown to lower postprandial lipoaemia in adolescents [13] and blood pressure in adults [11], it is pertinent to identify whether the favourable changes in glycaemic control, SBP and fat oxidation observed in the HIIE trial remain the following day.

5. CONCLUSION

This study investigated the influence of accumulating work-matched exercise of different intensities on postprandial plasma TAG, glucose, SBP and fat oxidation in male and female adolescents. We have demonstrated that repeating very brief bouts of HIIE, but not MIE, is effectual in lowering postprandial plasma glucose concentrations and SBP, and increasing resting fat oxidation on the same day in adolescents. These observations are encouraging, and add to a growing body of literature which identifies that accumulating short bouts of high-
intensity exercise may be more important for cardiometabolic health than MIE [15-17]. Further work is now needed to establish the efficacy of accumulating HIIE on parameters of postprandial health in adolescents with established cardiometabolic risk factors, and whether this translates into a clinically meaningful outcome with time.

Acknowledgments
We thank the staff and participants at Exmouth Community College, Devon, UK for their participation in this project.

Funding
This study was supported by the Sport and Health Sciences Research Committee, College of Life and Environmental Sciences, University of Exeter.

Disclosure statement
The authors confirm the absence of any conflicts of interest.

Author contributions
BB, AB, CW and NA designed the study. BB and AW conducted the research. BB, AW, SJ and AB analysed the data. BB and AB wrote the initial draft of the manuscript. All authors edited the manuscript and approved the final draft.
REFERENCES


FIGURE LEGENDS

**Figure 1** Protocol schematic. Numbers 1-3 represent either rest, moderate-intensity exercise or high-intensity interval exercise. Arrows represent capillary blood samples for plasma TAG and glucose concentrations; grey boxes represent the assessment of resting metabolic rate and blood pressure; HFM = high fat meal.

**Figure 2** Mean plasma TAG and glucose concentrations in the control (○), moderate-intensity exercise trial (▲) and high-intensity interval exercise trial (■) conditions for boys (A, C) and girls (B, D). The high fat meals are represented by the black rectangles. * = $P<0.05$ for HIIE vs CON. Error bars describe the standard deviation.

**Figure 3** Total area under the curves for postprandial systolic blood pressure (A) and fat oxidation (B) vs time (6 hours) collapsed for the boys and girls ($n=20$). CON, control trial; MIE, moderate-intensity exercise trial; HIIE, high-intensity interval exercise trial. * = $P<0.05$ for HIIE vs CON; # = $P<0.05$ for HIIE vs MIE. Error bars describe the standard deviation.
Fig. 2
Fig. 3
Table 1: Participant characteristics

<table>
<thead>
<tr>
<th></th>
<th>Boys (n = 9)</th>
<th>Girls (n = 10)</th>
<th>P value</th>
<th>Effect Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (y)</td>
<td>13.5 ± 0.3</td>
<td>13.9 ± 0.5</td>
<td>0.05</td>
<td>0.96</td>
</tr>
<tr>
<td>Body mass (kg)</td>
<td>59.4 ± 18.1</td>
<td>55.2 ± 7.7</td>
<td>0.51</td>
<td>0.31</td>
</tr>
<tr>
<td>Stature (m)</td>
<td>1.68 ± 0.14</td>
<td>1.63 ± 0.05</td>
<td>0.28</td>
<td>0.49</td>
</tr>
<tr>
<td>Percentage body fat (%)</td>
<td>18 ± 5</td>
<td>23 ± 7</td>
<td>0.06</td>
<td>0.96</td>
</tr>
<tr>
<td>$\dot{V}\text{O}_2\text{max}$ (L·min$^{-1}$)</td>
<td>2.60 ± 0.87</td>
<td>2.02 ± 0.27</td>
<td>0.09</td>
<td>0.92</td>
</tr>
<tr>
<td>$\dot{V}\text{O}_2\text{max}$ (mL·kg$^{-1}$·min$^{-1}$)</td>
<td>43.7 ± 6.0</td>
<td>36.8 ± 3.9</td>
<td>0.01</td>
<td>1.39</td>
</tr>
<tr>
<td>GET (L·min$^{-1}$)</td>
<td>1.29 ± 0.40</td>
<td>1.09 ± 0.18</td>
<td>0.20</td>
<td>0.66</td>
</tr>
<tr>
<td>GET (% $\dot{V}\text{O}_2\text{max}$)</td>
<td>50 ± 7</td>
<td>55 ± 9</td>
<td>0.27</td>
<td>0.52</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>108 ± 14</td>
<td>111 ± 10</td>
<td>0.50</td>
<td>0.32</td>
</tr>
<tr>
<td>HR (b·min$^{-1}$)</td>
<td>62 ± 8</td>
<td>68 ± 9</td>
<td>0.04</td>
<td>0.71</td>
</tr>
<tr>
<td>Glucose (mmol·L$^{-1}$)</td>
<td>5.17 ± 0.36</td>
<td>5.11 ± 0.32</td>
<td>0.72</td>
<td>0.15</td>
</tr>
<tr>
<td>TAG (mmol·L$^{-1}$)</td>
<td>0.23 ± 0.10</td>
<td>0.16 ± 0.09</td>
<td>0.07</td>
<td>0.73</td>
</tr>
</tbody>
</table>

$\dot{V}\text{O}_2$, oxygen uptake; GET, gas exchange threshold; SBP, systolic blood pressure; HR, heart rate; TAG, plasma triacylglycerol. SBP, HR, plasma [glucose] and plasma [TAG] are measured in the fasted state.
Data presented as mean ± SD for boys and girls.
Table 2: Accelerometer and food diary data during the 48 hours preceding each trial

<table>
<thead>
<tr>
<th></th>
<th>CON</th>
<th>MIE</th>
<th>HIIE</th>
<th>MIE vs. CON 95% CI</th>
<th>HIIE vs. CON 95% CI</th>
<th>HIIE vs. MIE 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moderate-vigorous activity (min day^{-1})</td>
<td>45 ± 16</td>
<td>44 ± 11</td>
<td>47 ± 20</td>
<td>-9 to 7</td>
<td>-7 to 12</td>
<td>-7 to 13</td>
</tr>
<tr>
<td>Total energy intake (kcal day^{-1})</td>
<td>1873 ± 520</td>
<td>1884 ± 487</td>
<td>1942 ± 498</td>
<td>-231 to 274</td>
<td>-397 to 452</td>
<td>-209 to 221</td>
</tr>
<tr>
<td>Energy from carbohydrates (%)</td>
<td>49 ± 5</td>
<td>47 ± 5</td>
<td>49 ± 5</td>
<td>-8 to 4</td>
<td>-5 to 4</td>
<td>-3 to 7</td>
</tr>
<tr>
<td>Energy from fat (%)</td>
<td>35 ± 6</td>
<td>35 ± 6</td>
<td>37 ± 5</td>
<td>-6 to 7</td>
<td>-4 to 8</td>
<td>-3 to 6</td>
</tr>
<tr>
<td>Energy from protein (%)</td>
<td>16 ± 4</td>
<td>17 ± 4</td>
<td>15 ± 3</td>
<td>-1 to 4</td>
<td>-5 to 2</td>
<td>-5 to -1</td>
</tr>
</tbody>
</table>

CON, control trial; MIE, moderate-intensity exercise trial; HIIE, high-intensity interval exercise trial
95% CI = 95% confidence limits for the true difference
Data have been pooled as ANOVA analysis revealed no main effect for sex
Table 3: Physiological and perceptual responses to MIE and HIIE

<table>
<thead>
<tr>
<th></th>
<th>MIE</th>
<th>HIIE</th>
<th><strong>P</strong> value</th>
<th>Effect Size</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Boys</strong>*</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean HR (b∙min(^{-1}))</td>
<td>130 ± 14</td>
<td>141 ± 9</td>
<td>0.01</td>
<td>0.81</td>
</tr>
<tr>
<td>Mean HR (% HR(_{\text{max}}))</td>
<td>67 ± 5</td>
<td>73 ± 3</td>
<td>0.01</td>
<td>0.81</td>
</tr>
<tr>
<td>Mean (\dot{V}O_2) (L∙min(^{-1}))</td>
<td>1.21 ± 0.25</td>
<td>1.45 ± 0.34</td>
<td>&lt;0.01</td>
<td>0.88</td>
</tr>
<tr>
<td>Mean (\dot{V}O_2) (% (\dot{V}O_{2\text{max}}))</td>
<td>48 ± 7</td>
<td>58 ± 7</td>
<td>&lt;0.01</td>
<td>0.93</td>
</tr>
<tr>
<td>RER</td>
<td>0.86 ± 0.04</td>
<td>1.01 ± 0.95</td>
<td>&lt;0.01</td>
<td>0.95</td>
</tr>
<tr>
<td>RPE</td>
<td>3 ± 1</td>
<td>6 ± 1</td>
<td>&lt;0.01</td>
<td>0.90</td>
</tr>
<tr>
<td>PACES</td>
<td>61 ± 14</td>
<td>66 ± 9</td>
<td>0.11</td>
<td>0.54</td>
</tr>
<tr>
<td>Work performed (kJ)</td>
<td>120 ± 28</td>
<td>120 ± 28</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Energy Expenditure (kJ)</td>
<td>220 ± 46</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><strong>Girls†</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean HR (b∙min(^{-1}))</td>
<td>135 ± 13</td>
<td>148 ± 10</td>
<td>0.01</td>
<td>0.83</td>
</tr>
<tr>
<td>Mean HR (% HR(_{\text{max}}))</td>
<td>71 ± 5</td>
<td>76 ± 5</td>
<td>0.01</td>
<td>0.91</td>
</tr>
<tr>
<td>Mean (\dot{V}O_2) (L∙min(^{-1}))</td>
<td>1.09 ± 0.14</td>
<td>1.25 ± 0.15</td>
<td>&lt;0.01</td>
<td>0.81</td>
</tr>
<tr>
<td>Mean (\dot{V}O_2) (% (\dot{V}O_{2\text{max}}))</td>
<td>54 ± 6</td>
<td>63 ± 7</td>
<td>&lt;0.01</td>
<td>0.81</td>
</tr>
<tr>
<td>RER</td>
<td>0.89 ± 0.03</td>
<td>1.04 ± 0.05</td>
<td>&lt;0.01</td>
<td>0.95</td>
</tr>
<tr>
<td>RPE</td>
<td>3 ± 1</td>
<td>5 ± 1</td>
<td>&lt;0.01</td>
<td>0.92</td>
</tr>
<tr>
<td>PACES</td>
<td>62 ± 11</td>
<td>58 ± 8</td>
<td>0.48</td>
<td>0.24</td>
</tr>
<tr>
<td>Work performed (kJ)</td>
<td>103 ± 9</td>
<td>103 ± 9</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Energy Expenditure (kJ)</td>
<td>172 ± 36</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

HR, heart rate; \(\dot{V}O_2\), oxygen uptake; MIE, moderate-intensity exercise trial; HIIE, high-intensity interval exercise trial.
Data presented as mean ± SD for MIE and HIIE.
* \(n = 10\) apart from mean HR where \(n = 9\) due to loss of telemetric data
† \(n = 10\) apart from mean HR where \(n = 8\) due to loss of telemetric data
Table 4: Postprandial observation of TAG and glucose

<table>
<thead>
<tr>
<th></th>
<th>CON</th>
<th>MIE</th>
<th>HIIE</th>
<th>MIE vs. CON 95% CI</th>
<th>HIIE vs. CON 95% CI</th>
<th>HIIE vs. MIE 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Boys (n = 9)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TAUC-TAG (mmol · L⁻¹ 6 h)</td>
<td>4.27 ± 1.53</td>
<td>4.54 ± 2.07</td>
<td>4.22 ± 1.11</td>
<td>-0.72 to 1.82</td>
<td>-0.69 to 0.88</td>
<td>-1.26 to 0.35</td>
</tr>
<tr>
<td>IAUC-TAG (mmol · L⁻¹ 6 h)</td>
<td>2.89 ± 1.40</td>
<td>3.28 ± 1.79</td>
<td>2.96 ± 0.99</td>
<td>-0.31 to 1.57</td>
<td>-0.49 to 0.83</td>
<td>-1.03 to 0.11</td>
</tr>
<tr>
<td><strong>Girls (n = 10)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TAUC-TAG (mmol · L⁻¹ 6 h)</td>
<td>2.94 ± 1.58</td>
<td>2.55 ± 0.98</td>
<td>2.79 ± 1.26</td>
<td>-1.53 to 0.74</td>
<td>-0.85 to 0.56</td>
<td>-0.48 to 0.96</td>
</tr>
<tr>
<td>IAUC-TAG (mmol · L⁻¹ 6 h)</td>
<td>1.94 ± 1.17</td>
<td>1.67 ± 0.78</td>
<td>1.77 ± 0.72</td>
<td>-1.11 to 0.57</td>
<td>-0.75 to 0.43</td>
<td>-0.40 to 0.61</td>
</tr>
<tr>
<td><strong>All (n = 19)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TAUC-glucose (mmol · L⁻¹ 6 h)</td>
<td>31.34 ± 1.93</td>
<td>31.26 ± 1.65</td>
<td>30.54 ± 1.83</td>
<td>-0.93 to 0.82</td>
<td>-1.64 to -0.08*</td>
<td>-1.57 to -0.04*</td>
</tr>
<tr>
<td>IAUC-glucose (mmol · L⁻¹ 6 h)</td>
<td>0.53 ± 1.88</td>
<td>0.54 ± 1.51</td>
<td>0.04 ± 1.71</td>
<td>-1.11 to 1.06</td>
<td>-1.50 to 0.49</td>
<td>-1.47 to 0.50</td>
</tr>
</tbody>
</table>

TAUC, total area under the curve; IAUC, incremental area under the curve; TAG, triglycerides; CON, control trial; MIE, moderate-intensity exercise trial; HIIE, high-intensity interval exercise trial
95% CI = 95% confidence limits for the true difference
Glucose data have been pooled as ANOVA analysis revealed no main effect for sex
* = P < 0.05