Original research

Effects of breaking up sitting on adolescents' postprandial glucose after consuming meals varying in energy: a cross-over randomised trial

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ABSTRACT

Objectives: To explore the impact of uninterrupted sitting versus sitting with resistance-type activity breaks on adolescents' postprandial glucose responses while consuming a diet varying in energy.
Design: Cross-over randomised trial.
Methods: Thirteen healthy participants (16.4 ± 1.3 years) completed a four-treatment cross-over trial: (1) uninterrupted sitting + high-energy diet; (2) sitting with breaks + high-energy diet; (3) uninterrupted sitting + standard-energy diet; and (4) sitting with breaks + standard-energy diet. For all four conditions, two identical meals were consumed; at 0 h and 3 h. A continuous glucose monitoring system (CGM) recorded interstitial glucose concentrations every five minutes. Linear mixed models examined differences in glucose positive incremental area under the curve (iAUC) and total AUC between the sitting and diet conditions for the first meal, second meal and entire trial period.
Results: Compared to the uninterrupted sitting conditions, the breaks condition elicited a 36.0 mmol/L/h (95%CI 6.6–65.5) and 35.9 mmol/L/h (95%CI 6.6–65.5) lower iAUC response after the first and second meal, respectively, but not for the entire trial period or for total AUC. Compared to the standard-energy diet, the high-energy diet elicited a 55.0 mmol/L/h (95%CI 25.8–84.2) and 75.7 mmol/L/h (95%CI 8.6–142.7) higher iAUC response after the first meal and entire trial, respectively. Similar response to the high-energy diet were observed for total AUC.
Conclusions: According to iAUC, interrupting sitting had a significant effect on lowering postprandial glucose for both dietary conditions, however, it was not significant when examining total AUC. Larger studies are needed to confirm these findings.
Clinical Trial Registration Number: ACTRN12615001145594.

1. Introduction

Chronic health conditions thought to develop during late adulthood, such as metabolic syndrome and type 2 diabetes, are now occurring at a younger age than in previous decades.[1] Some studies in youth,[2] but not all,[3] suggest that engaging in high volumes of sedentary time is associated with increased fasting blood glu-

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cose levels and insulin resistance which are key risk factors for developing these conditions. Thus, targeting reductions in sedentary time has the potential to improve an adolescent's metabolic profile and reduce their risk of developing metabolic syndrome or type 2 diabetes.

In the last decade, acute experimental trials have reported that interrupting sitting time with brief bouts of light- or moderate-intensity walking reduces postprandial glucose and insulin levels in healthy[4,5] and overweight/obese adults.[6,7] Moreover, two recent randomised cross-over studies have extended these findings into younger populations.[8,9] One study by Belcher et al. demonstrated that interrupting 3 h of continuous sitting with 3-min light-intensity walking breaks every 30 min reduced postprandial glucose by 7% and insulin levels by 32% in children aged 7–11 years.[8] However, the other study by Saunders et al. reported no significant differences in glucose or insulin responses in children aged 10–14 years when 8 h of prolonged sitting was interrupted with 2-min light- or moderate-intensity walking breaks every 20 min.[9]

The inconsistent findings between experimental studies may be explained in part by disparities in the energy content of the meals and by differences in blood sampling intervals. In the study by Saunders et al.,[9] although the meals were specifically developed for a young population, they may not have contained enough energy or the most suitable foods or beverages to raise glucose and insulin levels as has been demonstrated in adult studies.[4–6] In contrast, the findings by Belcher et al.[8] indicate that the use of meals containing a large amount of energy (e.g. a buffet style meal) may be necessary to elicit a sufficient glycaemic challenge to observe significant differences between intervention conditions. In addition, blood samples were collected at different time intervals (i.e. 30 min[8] versus 90 min[9]), which may not have been sufficiently regular to capture the fluctuations in glucose often seen in healthy, young populations. Continuous glucose monitoring systems (CGM) are minimally invasive and assesses interstitial glucose concentrations every 5 min. This provides the opportunity to assess the various temporal changes in blood glucose levels.[10] Although these devices are widely used clinically and in patients with type 1 or 2 diabetes,[10,11] no study to date has used a CGM to examine the acute effects of interrupting sitting on glucose responses in a healthy, young population.

Given the key role that dietary intake has on glucose metabolism,[12] it is surprising that no study to date has examined the effects of interrupting prolonged sitting on postprandial glucose when the dietary component is manipulated. Therefore, the aim of this pilot study was to explore the impact of uninterrupted sitting versus sitting with activity breaks on adolescents' postprandial glucose responses after consuming a diet varying in energy (e.g. high-energy versus standard-energy) across three time periods; (1) after the first meal; (2) after the second meal; (3) and the entire trial period. While we anticipated that the high-energy diet would elicit increased glucose responses overall, we hypothesised that the activity breaks would significantly attenuate postprandial glucose after the first meal, second meal and the entire trial period when compared to uninterrupted sitting for both diet conditions.

2. Methods

Ethical approval was obtained by Deakin University Human Research Ethics Committee in April 2015 (#2015-039). Study participants were recruited between September and December 2015 via flyers, local newspaper advertisement and word of mouth in Melbourne, Australia. Eligible participants were aged between 14 and 17 years, in good general health, not on any glucose lowering medications and had no dietary allergies. Eligibility criteria were assessed via phone screening interview with the participant or

their parent. The recruitment stages are shown in Fig. S1. Written parental and participant consent was provided by 16 participants, with 15 participants completing the four trial conditions over the school summer holidays (November 2015 to January 2016). Participants who completed all four conditions received a FitBit® as compensation for their time.

The study was an acute, cross-over factorial randomised trial involving four experimental conditions each performed over a 6-h period: (1) uninterrupted sitting and high-energy diet; (2) interrupted sitting with 2-min activity breaks every 18-min and high-energy diet; (3) uninterrupted sitting and standard-energy diet; and (4) interrupted sitting with 2-min activity breaks every 18-min and standard-energy diet. For each condition, participants received two meals, consumed at 0 and 3 h. The 2-min activity breaks involved body-weight resistance exercises and included 30-s half squats, 30-s calf raises, 30-s knee lifts and 30-s step-ups. Resistance-type activities were selected as resistance-type activities utilises larger muscle groups,[13] and thus may promote an increased energy expenditure and higher glucose uptake. The study protocol and an example of one of the experimental conditions is shown in Fig. 1.

After enrolment, each participant was assigned one of the pre-determined condition orders using a sequence which was randomly computer-generated by a research assistant. Participants were kept blinded to the order in which they completed each condition until the morning of each visit. Since moderate-intensity physical activity has been shown to have no residual effects on plasma glucose past a 17-h period,[14] a one-day minimum washout period was selected between trial conditions. The 24-h prior to each condition, participants were asked to refrain from participating in any moderate-to-vigorous physical activity (MVPA). Compliance to MVPA was self-reported using a checklist completed the morning on each condition day.

For all four experimental conditions, participants arrived at the laboratory between 0730 and 0800 after a 12-h overnight fast. At the first visit only, participants' height, weight, waist circumference, and blood pressure were measured using standard procedures.[15,16] Participants had access to a television, DVDs, sedentary video games, and computer and internet services. Participants were provided with a comfortable chair throughout the trial and were instructed to sit upright and minimise any movement when not performing the activity breaks. If participants needed to use the lavatory, they were escorted in a wheelchair. Research staff directly supervised the participants at all times to ensure compliance with the protocol. For descriptive purposes, participants completed a questionnaire about their usual MVPA[17] and sedentary behaviours levels[18] on the first condition day, and completed a 1-day food diary of food, beverages and medications consumed the day before each of the four experimental conditions. The food diaries were collected by researchers the morning of each condition to ensure compliance to the 12-h overnight fast and to calculate usual energy and macronutrient intakes.

Prior to the first and third condition, two researchers visited the participant's home to insert a CGM (Medtronic iPro2™, Northridge, USA) on the right side of the participant's lower back. After the CGM was inserted, an adhesive, waterproof strip was placed over the top of the CGM to minimise movement of the sensor and to allow participants to shower. Due to the CGM recorder having a battery life of seven days, a new sensor was inserted prior to the third trial condition to be worn during the final two conditions. To calibrate the CGM, two capillary blood glucose samples were taken from each participant by the researchers during the two home visits, and three during the trial days. The capillary blood samples were used to calibrate the CGM at the time of downloading the data. At the home visits, participants also received four standardised dinners to consume the night prior to each of the trial conditions. The
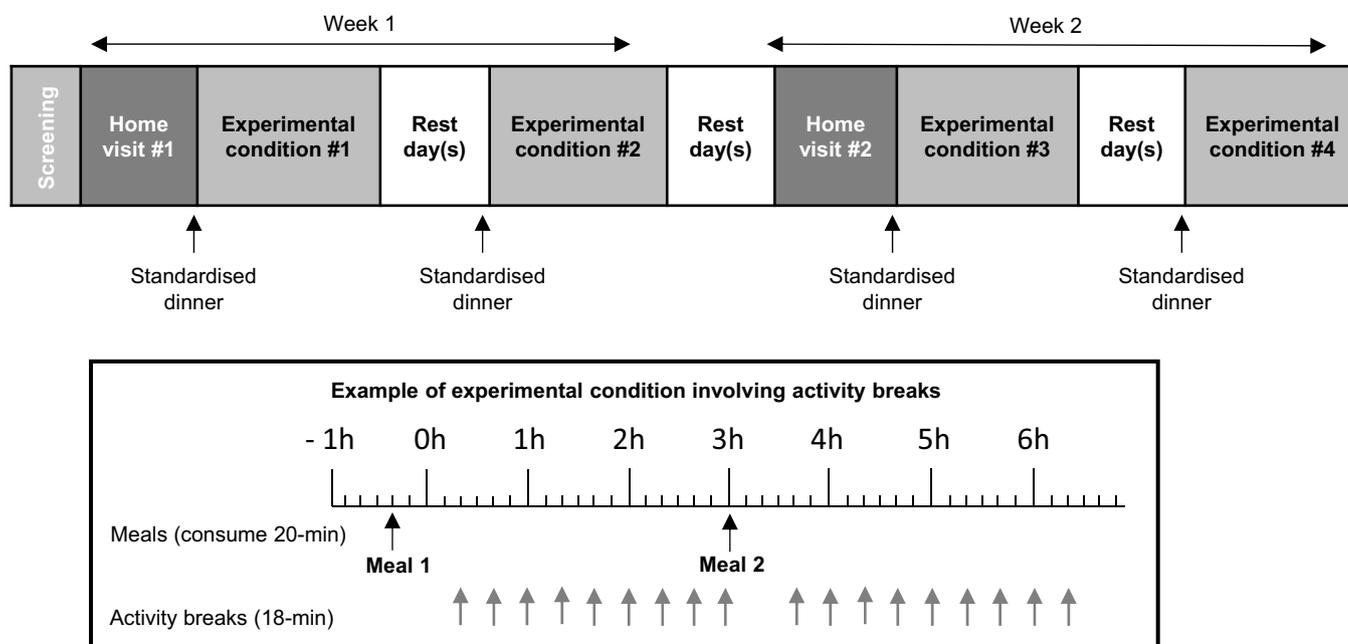
Fig. 1. Study design and experimental protocol.

standardised dinner contained 2874 kj and included a 400 g frozen lasagne, 30 g wholemeal bread and 50 g of mixed vegetables. Participants were instructed to consume the entire meal at 1900, within 20 min and not to consume any other food or beverages except for water. Compliance to the dinner protocol was reported to the researchers the following morning.

The contents of the high-energy and standard-energy diets are described in the online supplement file (Table S1). During each of the trial conditions, participants received two high-energy meals or two standard-energy meals; the first meal was consumed at the start of the trial (between 0730 and 0830) and the second meal (exactly the same as the first meal) was provided 3 h after ingestion of the first meal. The energy contents of the meals were based on the estimated-energy requirements for an adolescent aged 17 years with a body mass index (BMI) of 25 kg/m^2 (averaged for females and males).[19] In order to assess the effects of energy intake (kilojoules; kj), both the high-energy and standard-energy diets contained similar macronutrient composition with the high-energy diet containing 70% more kilojoules than the standard-energy diet. The kilojoules and macronutrient contents of the diets were calculated using dietary analysis software (FoodWorks Version 7, Xyris Software Pty Ltd, Australia) based on standard Australian food composition tables.[20] Participants were instructed to consume the entire meal within 20 min. Any food or beverage left over was weighed and recorded and used to calculate the average percentage of kilojoules consumed during each experimental condition for each participant.

Information regarding the analyses on the sequences of the cross-over design and interaction effects can be found in the online supplement file. As this study was a pilot study, it was estimated that 16 participants (equates to 64 conditions completed) would be sufficient to determine the pragmatic considerations of examining an adolescent population. Using the CGM data, the primary outcome was positive incremental area under the curve (iAUC) and was calculated using the trapezoidal method[21] (GraphPad Prism v6) across three time periods: (1) three-hours post first meal (includes eating time), using the baseline fasting glucose just prior to the consumption of the first meal as the reference point; (2) three-hours post second meal (includes eating time), using the glucose value just prior to consuming the second meal as baseline; and

(3) the entire trial time (including both the first and second meal responses), using the baseline fasting glucose just prior to the consumption of the first meal as the reference point. A secondary outcome was to examine total AUC. Similar to iAUC, total AUC was calculated using the trapezoidal method across the same three time periods, however, zero was used as the reference point for all time periods.

Linear mixed models were used to examine the effect of the sitting and diet conditions on iAUC and total AUC with period effect (i.e. the effect of the order at which the participant received the condition), sitting/breaks and diet conditions as fixed effects, and participant as a random effect. The interaction between the sitting/breaks and diet conditions was not examined as the pilot study was not powered to detect this. To account for potential differences in the percentage of meals consumed and fasting baseline glucose at each condition, these variables were adjusted for in the analyses. Pseudo effect size measure (Cohen's D) were calculated using the EffectSizeMixed SAS macro.[22] Post-hoc power calculations were performed, with results in the online supplement file. Analyses were conducted in SAS version 9.3 (SAS Institute, Cary, NC) and $p < 0.05$ was considered significant.

3. Results

Out of the 16 participants who were randomised, 15 participants completed the four experimental conditions. Two participants were excluded from the analysis due to non-compliance to the study protocol, leaving 13 participants with complete data for the analyses. The baseline characteristics of the 13 participants are outlined in the online supplement file (Table S2). Overall, the mean age of participants was 16.4 ± 1.3 years (39% male) and BMI was 20.6 ± 2.5 kg/m^2. On average, participants spent 428 ± 144 min/day in self-reported sedentary behaviour and 43 ± 44 min/day in self-reported MVPA.

Fig. 2 summarises mean glucose responses across the four conditions and Table 1 summarises the AUCs responses across the four conditions and differences in the sitting/breaks and diet effects. Compared to the sitting conditions, the breaks conditions elicited a 36.0 mmol/L/h (95%CI 6.6–65.5) and 35.9 mmol/L/h (95%CI 8.6–63.1) significantly lower iAUC response after the first
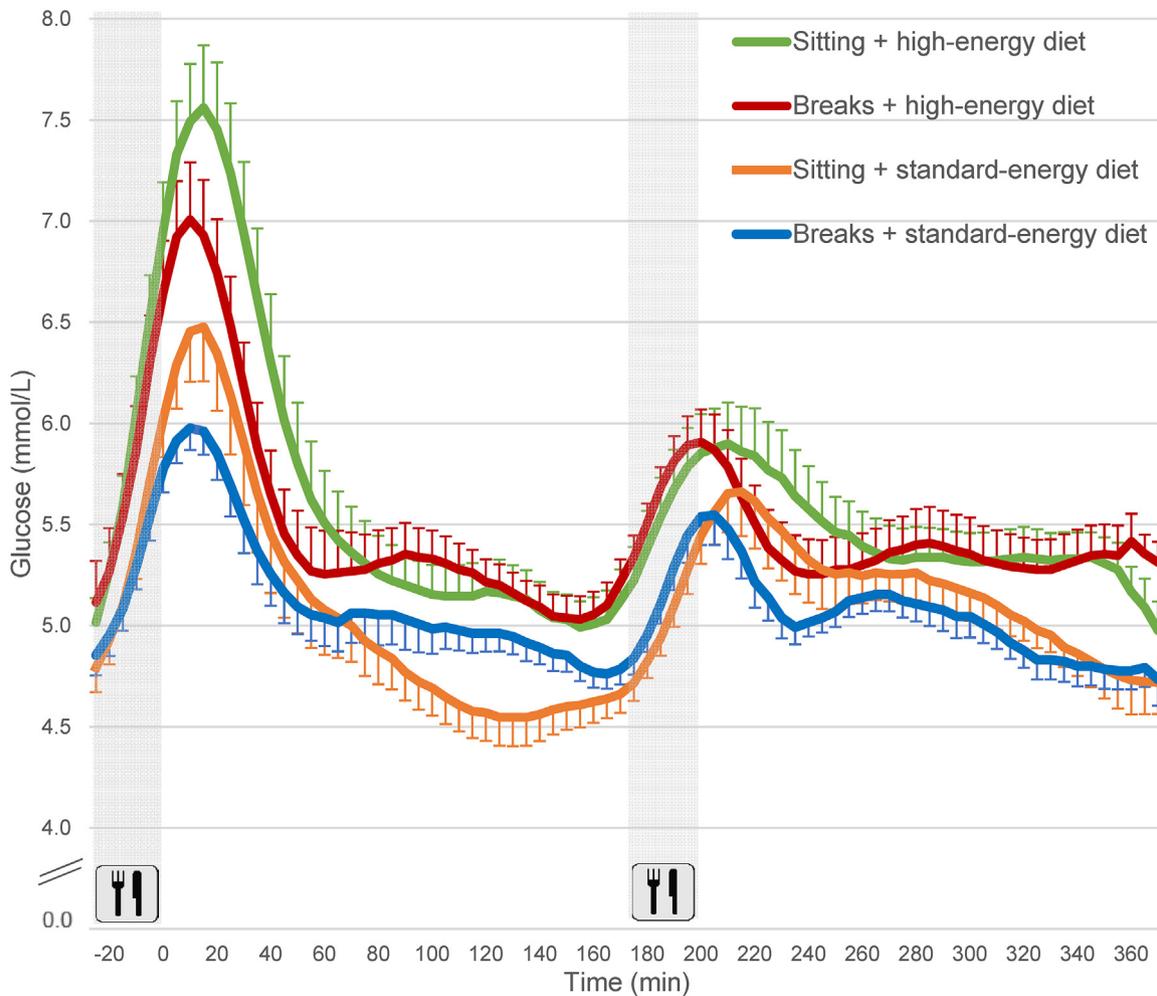
Fig. 2. Mean (SE) postprandial glucose responses across the four conditions.

Table 1
Mean ± SE area under the curve (AUC) for all four conditions and differences (95%CI) in the effects of the sitting conditions and diet conditions.

	Mean (SE) AUCs for each condition[a]				Sitting versus breaks[b]		High-energy versus standard-energy[b]	
	(1) High-energy diet + sitting	(2) High-energy diet + breaks	(3) Standard-energy diet + sitting	(4) Standard-energy diet + breaks	Difference (95%CI)	Effect size	Difference (95%CI)	Effect size
iAUC (mmol/L/h)								
First meal	166.6 (21.8)	115.5 (19.1)	93.0 (14.1)	75.1 (12.7)	36.0 (6.6, 65.5)[*]	0.7	55.0 (25.8, 84.2)[**]	1.1
Second meal	105.1 (13.6)	84.3 (16.1)	116.4 (22.4)	70.9 (10.1)	35.9 (8.6, 63.1)[*]	0.8	2.5 (−25.3, 30.2)	0.1
Entire period	262.6 (41.4)	207.6 (48.2)	175.3 (30.1)	132.7 (24.4)	46.8 (−19.7, 113.4)	0.4	75.7 (8.6, 142.7)[*]	0.7
Total AUC (mmol/L/h)								
First meal	1145.4 (27.4)	1114.5 (27.3)	1021.7 (27.9)	1029.0 (27.5)	12.8 (−24.9, 50.4)	0.2	103.8 (66.6, 141.1)[**]	1.6
Second meal	1093.5 (23.0)	1084.9 (22.7)	1026.1 (23.5)	1004.6 (23.0)	30.9 (−36.4, 98.1)	0.2	78.2 (36.3, 112.1)[**]	1.1
Entire period	2205.0 (46.4)	2164.9 (46.1)	2037.8 (47.3)	2017.1 (46.5)	14.8 (−22.4, 51.9)	0.3	157.1 (89.1, 225.1)[**]	1.3

[a] Mean and standard error (SE) for each condition estimated under a linear mixed model including participant as a random effect and period, diet, breaks, interaction diet/breaks as fixed effects.

[b] Sitting/breaks and high-energy/standard energy diet mean differences (95% CI) estimated under the same model, excluding the interaction term which was not significant in the former model.

[*] p < 0.05.

[**] p < 0.001.

and second meal, respectively. However, this was no longer significant when examining iAUC across the entire trial period, nor for total AUC for any of the three time periods. For both the iAUC and total AUC, the breaks conditions had a moderate/high (0.4–0.7) and small effect (0.2–0.3), respectively, on lowering postprandial glucose. Relative to the standard-energy diet, the high-energy diet elicited a 55.0 mmol/L/h (95%CI 25.8–84.2) and 75.7 mmol/L/h (95%CI 8.6–142.7) higher iAUC response after the first meal and entire trial, respectively. No significant diet effects on iAUC were observed for the second meal. Similar responses to the high-energy meal were observed for total AUC, with large effect sizes observed (1.1–1.6).

4. Discussion

This is the first study to explore the impact of uninterrupted sitting and breaking up sitting with resistance-type activity breaks on adolescents' postprandial glucose while consuming a diet varying in energy. The findings showed that when examining the first and second meal responses separately, the activity breaks significantly attenuated the postprandial iAUC responses after both the first and second meal for both diet conditions, but did not significantly attenuate iAUC for the entire trial period, nor for when total AUC was examined. In addition, relative to the standard-energy diet, the study found that the high-energy diet elicit a higher postprandial glucose response for both iAUC and total AUC for both the sitting and breaks conditions.

Previous studies in children[8] and adults[4–7] have shown interrupting sitting time has beneficial metabolic effects on glycemic control. However, this is the first study to examine, whether regularly interrupting sitting time with activity breaks has beneficial effects on postprandial iAUC after consuming both high-energy and standard-energy diets. Interestingly, activity breaks significantly lowered iAUC but not total AUC. Although iAUC is most commonly used to assess acute glucose responses to meals relative to baseline (or fasting) glucose values, it excludes data that drop below baseline. We therefore calculated both iAUC and total AUC to provide a more comprehensive picture of the glycaemic responses. The varied effects sizes for these two glucose metrics is likely attributable to the different techniques involved in their calculation, but highlights the potential importance of considering which glucose parameters are examined – particularly across consecutive meals – and their physiological relevance.

In addition, while the measures used in this study do not permit conclusions about the potential mechanisms responsible for the reductions in postprandial glucose for iAUC, increases in contraction-mediated glucose uptake as a result of the activity breaks may play an important role.[23] As the activity breaks in the current study involved simple resistance exercises that can be performed with minimal equipment, these activity breaks could be easily incorporated into pedagogical approaches or even facilitated via environmental changes (e.g. height adjustable desks) in schools[24] and/or in the adolescents' home.

The study also observed, compared to the standard-energy diet, consuming a high-energy diet resulted in a higher postprandial glucose response (for both iAUC and total AUC) across the entire observational period, for both the uninterrupted sitting and breaks conditions. This is not surprising, given carbohydrates play a vital role in glucose metabolism[25] and the high-energy diet in the study contained 70% more kilojoules (and thus a greater amount of total carbohydrates) than the standard-energy meal. However, when examining the first and second meal responses separately, the study observed a significant higher postprandial glucose response for iAUC after the first meal, but not the second meal. This suggests that the first meal of the day (e.g. breakfast) may be the biggest contributor in terms of daily glycaemic responses and that glucose tolerance may improve for the subsequent meals later in the day (e.g. lunch and dinner).[26,27] Thus, future interventions could encourage adolescents to engage in physical activity soon after consuming breakfast (e.g. by walking to school) in order to have the most beneficial effects on postprandial glucose.

A strength of this study was the examination of four conditions performed over four separate days which allowed us to explore both effects of breaking up sitting and varying the energy content of the meals on postprandial glucose responses. Another key strength was examining an adolescent population, which, to date has been an understudied population group within the sedentary behaviour field. In addition, the use of the CGM among adolescent participants was able to capture a more complete portrayal of the postprandial glucose response (e.g. every 5 min), as opposed to collecting blood samples at hourly intervals. Lastly, the meals provided to the participants were based on whole foods, not supplements, commonly consumed by this population group.

Limitations include not measuring other glycaemic markers such as insulin, thus, limiting the ability to interpret the entire glycaemic response. Future studies using a continuous measure of glucose should also consider measuring key hormones including plasma insulin and C-peptide in order to examine potential mechanisms related to glucose-insulin kinetics. In addition, the study included both males and females in pre- and post-pubertal stages. Given the well-known effects that puberty has on glucose metabolism,[28] this may have increased the variability in postprandial glucose responses within and between participants. Further, the study only examined the acute glucose responses, therefore the current results cannot be extrapolated to long-term exposures. Lastly, the study was limited by a small sample size and it is unclear whether the findings would be exaggerated in youth who are overweight or obese. Thus, future studies are needed involving a larger sample size and involving overweight or obese adolescents to test the protocol in an at-risk sample.

5. Conclusion

The findings from the study demonstrated that compared to prolonged sitting, regularly interrupting sitting with simple resistance activity breaks after the first and second meals attenuated postprandial iAUC responses for both the high-energy and standard-energy diets. However, no significant breaks effects were observed for total AUC. Longer-term intervention studies are needed to confirm these findings, as well as studies examining interactions between the sitting and diet conditions in relation to cardiometabolic health risk factors.

Practical implications

- Interrupting sitting time with simple resistance activity breaks lowers incremental postprandial glucose responses to both high-energy and standard-energy diets. This suggests that breaking up prolonged sitting, particularly after consuming main meals, may be an effective intervention strategy for adolescents.
- Compared to the standard-energy diet, the high-energy diet elicited a higher positive incremental postprandial glucose response after the first meal, but not the second meal. This suggests the first meal of the day may be the biggest contributor in raising postprandial glucose levels.
- Larger and longer-term interventions are needed to confirm these findings, as well as studies examining interactions between the sitting and diet conditions in relation to cardiometabolic health risk factors.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at 10.1016/j.jsams.2017.06.002.

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