ORIGINAL ARTICLE

Interrupted sitting at work can alter postprandial glucose, triglycerides and non-esterified fatty acids (NEFA)

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ABSTRACT

Introduction: Prolonged sitting at work is a major health issue as sedentary behaviour has been linked to cardio-metabolic risks. Workplace interventions have trialled intermittent interruption to sitting throughout the working day to mitigate effects on employees' health. This study tested effects of the intensity of the interruptions to sitting on postprandial glucose, triglycerides and NEFA in a laboratory setting.

Materials and Methods: Overweight and obese adults (n=24) were recruited for a randomised cross over trial comparing, 1) uninterrupted sitting, 2) sitting interrupted by light intensity walking at 3 km.hr-1, and 3) vigorous intensity stair climbing at 60 steps.min-1. The interruptions to sitting were performed for two-minute bouts every 20 minutes following a meal. The moderate fat meal was provided two hours after the trial started and blood samples were collected each hour throughout the seven-hour sessions.

Results: There was a lower postprandial peak in plasma glucose and attenuation of the NEFA reductions for stair climbing compared to uninterrupted sitting. In contrast, triglycerides were reduced postprandially following light intensity walking compared to uninterrupted sitting.

Conclusion: Interrupted sitting with higher intensity demonstrates greater improvements postprandially.

KEYWORDS:
Interrupted sitting, light intensity walking, stair climbing, postprandial glucose, lipids

INTRODUCTION

Adults can spend up to half of their waking hour sitting with sitting in the workplace making a major contribution to this total.[1,2] Prolonged sitting is associated with increased cardio-metabolic risk; increases in BMI and waist circumference, impaired levels of high density lipoprotein cholesterol (HDL-C), elevated triglycerides and postprandial glucose in the circulation are associated with sedentary behaviour.[3-5] Many public health agencies have developed physical activity and sedentary behaviour guidelines to discourage prolonged sitting.[6,7] The recommendations are meant not only to reduce the total amount of sitting but also to regularly interrupt the prolonged sitting.[8] In epidemiological data, the duration of objectively measured sitting and the number of interruptions have been independently associated with cardio-metabolic risk.[9,10]

Physical activity performed in short bouts to interrupt sitting, regularly incorporated into the daily routine, has been shown to be more effective than a single equivalent session of continuous physical activity.[11-13] Since the main barrier to promoting sedentary breaks among employees may be a lack of work flexibility,[14] frequent short sedentary breaks might avoid disruption of office tasks and increase the feasibility of interventions while at work. Interrupted sitting interventions have been conducted in adults who were healthy and of healthy weight,[13,15-18] those who were sedentary and inactive[19,20] and those who were overweight and obese.[5, 21-22] Individuals receive a standard 'meal' in liquid form after two hours of interrupted sitting, with any physical activity interruptions beginning in sometime after the meal. As a result, these studies aim to assess the effects of interruptions by physical activity with differing levels of intensity on postprandial metabolism of glucose, triglycerides and NEFAs; since the postprandial increases in the concentrations of glucose, triglycerides and NEFAs in the bloodstream will contribute the most predictive power towards cardiovascular disease risk.[23]

For standing, the activity trialled with public health interventions that use 'sit-to-stand desks', Henson et al. reported reduced AUC following the meal for glucose and insulin[22] whereas neither Pulsford et al. nor Bailey & Locke reported any differences from sitting for glucose and insulin or glucose respectively.[16,20] In a 27-hour protocol, effects of intermittent standing after the first meal on glucose but not insulin were reported whereas an interruption of 30 minutes of moderate intensity walking reduced both glucose and insulin in the same period.[24] It is noteworthy that the Henson and co-workers sample were overweight and obese women with impaired glucose metabolism at baseline whereas the null findings for standing employed healthy individuals.[22] Consistent with this potential effects of the sample, Thorp et al. reported reductions in glucose AUC after five days of intermittent standing in a simulated office environment for overweight and obese participants on the fifth day, though there were no effects on insulin.[25] For light intensity walking, Bailey and Locke (3.2 km.hr-1) reported reductions in glucose whereas Pulsford et al. (3.6 km.hr-1), McCarthy et al. (3.0 km.hr-1) and Henson et al. (individually selected light

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intensity) reported reductions in both glucose and insulin consistent with the results in Dunstan and co-workers' original study.[5,16,19,20,22]

Exploratory analyses of the Australian group's overweight samples with consistent methodology revealed that greater intensity of physical activity performed during the interrupted would produce greater effects in the reduction of cardio-metabolic risk.[28] Such a result was consistent with the greater effects on the reduction of cardio-metabolic risk by more vigorous physical activity when compared to that by less intense physical activity, as shown by most of the available epidemiological data.[29-31]

Stair climbing requires more energy per minute than jogging, having been estimated as 9.6 metabolic equivalents (METs) of the sedentary state in the field[32] and 8.6 METs under laboratory conditions.[33] As such stair climbing is a vigorous physical activity. Experimental studies reveal that increased stair climbing can have effects on some cardio-metabolic risk factors.[34-39] To date, however, the effects of stair climbing have not been investigated as an interruption to sitting. The current study aimed to compare the acute effects of prolonged uninterrupted sitting with light intensity walking and the vigorous intensity physical activity of stair climbing on postprandial glucose, triglycerides (TG) and non-esterified fatty acid (NEFA). NEFA is free fatty acid (FFA) derived from the hydrolysis of fat or triglyceride in adipose tissue.[40]

MATERIALS AND METHODS

Study site

The intervention was conducted in the Stair Laboratory of the School of Sports, Exercise & Rehabilitation Sciences at the University of Birmingham.

Study population

Volunteers were recruited from a population within one-mile radius of the university. Posters were placed on the notice boards in the university buildings as well as distributed outside the campus adjacent to the university entrance to the pedestrians. Inclusion criteria involved: 1) overweight/obese, 2) healthy and not on any medication, 3) no knee or joint problem, 4) no restrictions on movement, and 5) sedentary (less than 60 min.wk-1 of MVPA). The exclusion criteria were; 1) pregnant, and 2) physically active.

Study design

This intervention was a randomised, counterbalanced cross-over design. The total number of participants was 24.

Sample Size Calculation

The required sample size was calculated using the two-means formula, with a power of 80% and a significance level (α) set at $p < 0.05$. This calculation considered several independent variables identified from previous literature, and after accounting for a 10% non-response rate, the final sample size was determined to be 24 participants.

Study protocol

The information sheet was emailed to the volunteers to allow them to fully understand the details of the study. They were also asked to complete physical activity readiness (PAR-Q) and international physical activity (IPA-Q) questionnaires to ensure eligibility and safety for participation. Those who were found eligible and agreed to join this study were given a consent form. Prior to the starting of the trials, participants were randomly assigned into one of eight groups. Each group was arranged to complete three sessions in a counterbalanced order involving the three different physical activities; uninterrupted sitting, walking and stair climbing. A minimum of six days of wash out period was kept between the sessions. Participants fasted overnight and refrained from drinks containing caffeine on the day of testing. Prior to the testing protocol, a baseline session was conducted to measure weight, height and BMI of participants.

a) Testing session

The testing session started at 9 a.m. with hourly withdrawal of 5 ml of fasting blood sample (-2h, -1h & 0h). After sitting for two hours, participants were provided with a moderate fat meal containing 35% fat, 47% carbohydrate and 18% protein providing 68.6 kJ energy per kg body mass as the British diet is moderate in fat. Calculation of total energy was based on participant's weight. The moderate fat meal comprised of a drink containing skimmed milk, single cream, sugar and flavoured yogurt, along with an energy bar, veggie snacks and fruit. The meal was consumed within 20 minutes before starting the first interruption with physical activity. Blood was collected every hour after the meal and the catheter was flushed every 15 minutes to avoid coagulation. Blood was transferred into red top vacutainer containing clot activator and left 30 minutes in room temperature to allow clotting. Samples were then centrifuged at 3000 rpm for 15 minutes at 4˚ C and kept in a -20˚ C freezer prior to analysis.

Each day's session involved three participants. One participant of the group completed sitting uninterrupted while the others either walked on the treadmill or completed stair climbing as interruptions to sitting. The uninterrupted sitting participant remained seated throughout the seven-hour period. The participant for interrupted sitting with light intensity activity walked on a treadmill at 3.0 km.hr-1 for two minutes and for the interrupted sitting by MVPA, participant performed stair climbing using the StairMaster 7000 at 60 steps.min-1 for two minutes. Walking at this speed requires approximately 2 METs. Climbing at this speed required 8.7 ± 0.6 METs in a young, aerobically fit sample, similar to the expenditure of 8.6 and 8.8 METs reported by Bassett et al. and Jetté et al. respectively.[33]

b) Blood samples analyses

Serum analyses were performed in duplicate using semi-automatic spectrophotometric clinical chemistry analyser, Instrumentation Laboratory (ILab 650, Cheshire, UK) to measure the concentrated glucose, triglycerides (TG) and non-esterified fatty acid (NEFA).

Ethical approval

Ethics for this study was approved by the Science, Technology, Engineering and Mathematics Ethical Review Committee, University of Birmingham with ethics reference number ERN_16_0472.

Data deduction and statistical analysis
To evaluate the changes of the variables in response to the test meal, the area under the curve (AUC) was calculated using the trapezoidal formula.

To allow comparison with previous research,5 AUC for two hours postprandial as well as AUC for the five-hour period were calculated. In addition, many metabolic processes when plotted over time are characterised by relatively simple shapes. To test for differences in these shapes between conditions, orthogonal polynomial trends across time were computed for the linear, quadratic, and cubic components, and these polynomial trends were analysed.

Data were analysed using the SPSS statistical package for Windows, version 20.0 (IBM Corp., Armonk, NY, USA). All participants provided complete data for the variables of interest; no missing data were observed. Shapiro-wilk's test for normality showed p>0.05 and visual judgment of the histogram, Q-Q plots and boxplots showed that the data were approximately normally distributed with skewness within the acceptable range of ± 1.96. A natural log transformation was applied to TG levels to improve the normality of their distribution. Repeated measures ANOVA with orthogonal polynomial contrasts was used to examine condition differences.

RESULTS

Total of 24 participants involved in the study had a BMI of 31.06 (SD=3.96) in obese class I (WHO, 2000). All participants would be classified as overweight (BMI > 25).

Glucose
Figure 1 below displays the pattern of glucose concentrations at one hour intervals throughout the seven-hour sessions. As can be seen, glucose increased immediately after meal and reached its peak at two hours postprandially.

To reiterate, an ordered relationship across conditions was predicted such that uninterrupted sitting > light walking > stair climbing based on the different intensities of the activities. The test of this ordered relationship would be the linear polynomial across conditions.

Analysis of the data from the immediate pre-meal values to five hours later, revealed a main effect of time (F(3, 60.8) = 51.92, p<0.001) that was described primarily by significant quadratic (F(1, 23) = 103.80, p<0.001; 63.6% of the variance) and cubic components (F(1, 23) = 65.74, p<0.001; 32.3% of the variance), with a significant interaction between the quadratic component and condition (F(1, 23) = 5.05, p =0.04). Follow-up analyses revealed differences in the quadratic component between uninterrupted sitting and stair climbing (p = .04) reflecting the lower peak response with stair climbing. There were no differences between the conditions for the cubic component over time (F(1, 23) = 0, 94 p=0.34). In addition, the omnibus ANOVA contained a significant linear component over time (F(1, 23) = 6.97, p=0.02; 2.7% of the variance) which did not differ between conditions (F(1, 23) = 2.13, p=0.16).

Table II summarises the AUC for glucose, NEFA and TG measured for the conditions of uninterrupted sitting, walking and stair climbing. As NEFA yielded a negative AUC due to the postprandial decrease, a change in concentration was calculated and subtracted from the level just before the meal to test the reductions postprandially. The actual level is shown in the table II.

For analyses of glucose AUC, a marginally significant linear polynomial over conditions (F(1, 23) = 3.22, p=0.086) was suggestive of an ordered relationship between the means for the five-hour period. Follow-up analyses suggested a smaller AUC for stair climbing than uninterrupted sitting (p=0.09) which would be significant with one tailed probability for the predicted effect. Similarly, a marginally significant linear polynomial for the two-hour AUC (F(1, 23) = 4.14, p = .054) was consistent with a smaller AUC for stair climbing than uninterrupted sitting (p = .06).

Triglycerides
Figure 2 displays the pattern for TG concentrations at one hour intervals throughout the seven-hour sessions. As can be seen, there was a steady increase postprandially in all three conditions.

Analysis of the immediate pre-meal and post-meal values revealed a main effect of time (F(2, 51.5) = 177.65, p<0.001) that was described primarily by a significant linear component (F(1, 23) = 297.62, p<0.001; 93.4% of the variance), with a suggestion that this differed between conditions (F(1, 23) = 3.13, p=0.09). The response also contained significant quadratic (F(1, 23) = 40.00, p<0.001; 5.1% of the variance) and cubic components (F(1, 23) = 16.95, p<0.001; 1.0% of the variance) which did not differ between conditions (both p>0.37). In addition, the ANOVA contained a main effect of condition (F(2, 41.6) = 3.99, p=0.03). In follow-up analyses, there was a smaller response for light walking than uninterrupted sitting (p=0.02), with a suggestion of a comparable difference for stair climbing (p=0.09).

For analyses of the TG AUC, a significant main effect of condition (F(2, 46) = 4.08, p=0.024) was accompanied by a marginally significant linear polynomial (F(1, 23) = 3.28, p=0.083), suggestive of an ordered relationship between the means for the five-hour period. Follow-up analyses revealed smaller magnitude AUC for light walking than sitting (p=0.02) with a suggestion of a similar difference with stair climbing (p=0.08). For the two-hour AUC, a significant main effect of condition (F(2, 43.5) = 3.59, p=0.04) was accompanied by a significant linear polynomial across conditions (F(1, 23) = 7.16, p=0.01). Follow-up of the main effect of condition revealed that light walking differed from uninterrupted sitting (p=0.04).

NEFA
Figure 3 displays the pattern for NEFA concentrations at one hour intervals throughout the seven-hour sessions. For NEFA, the level dropped postprandially as expected, with a nadir reached two hours after the meal.

Table I: Participants' characteristics and values of metabolites prior to the test meal (n=24)

Variable	Mean ± SD	Mean ± SD (mmol.L-1.hr-1)		
		Sitting uninterrupted	Walking	Stair climbing
Age	32.17 (5.57)			
Weight (kg)	85.34 (14.52)			
Height (cm)	165.05 (9.09)			
BMI	31.06 (3.96)			
Glucose		4.38 (0.55)	4.26 (0.38)	4.23 (0.32)
TG		1.32 (0.69)	1.19 (0.78)	1.22 (0.58)
NEFA		0.84 (0.33)	0.79 (0.21)	0.78 (0.23)

As shown in Table I above, there were no differences found between the conditions pre-meal (all p > .12). The accepted statistically significant p-value was <0.05 (two-tailed).

Table II: Area under the curve for glucose, NEFA and TG (n=24)

Variable	Mean ± SD (mmol.L-1)		
	Sitting uninterrupted	Walking	Stair climbing
2-hour test			
Glucose	5.65 (1.40)	5.44 (0.82)	5.30 (0.52)
TG	1.64 (0.87)	1.45 a (0.89)	1.46 (0.63)
NEFA	0.42 (0.15)	0.44 (0.017	0.38 (0.11)
5-h test			
Glucose	5.41 (0.99)	5.24 (0.52)	5.12 (0.44)
TG	2.25 (1.11)	1.95 [a] (1.21)	2.01 (0.94)
NEFA	0.28 (0.10)	0.32 (0.12)	0.30 (0.09)

[a] significantly different from uninterrupted sitting.

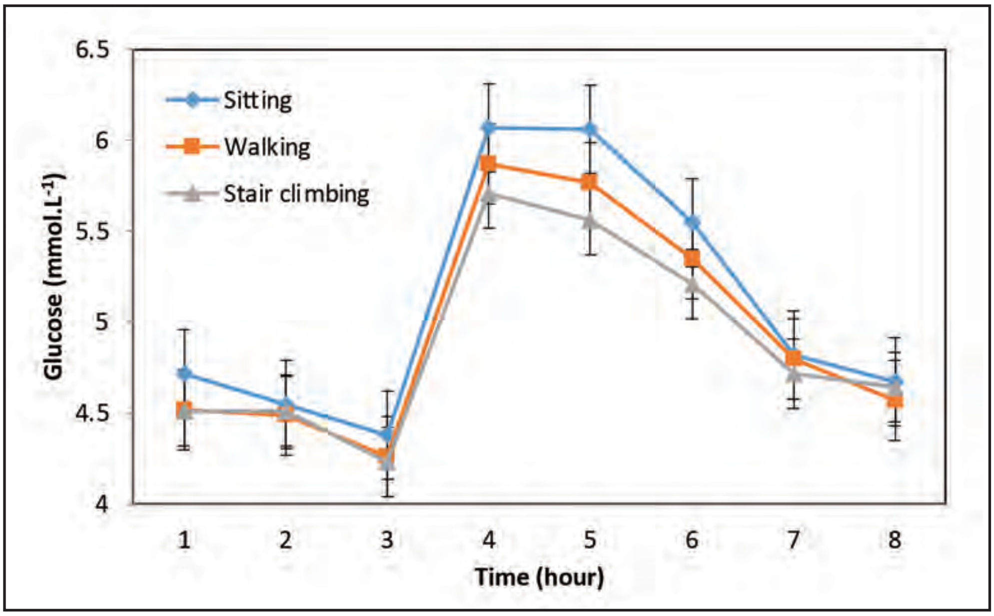

Fig. 1: Glucose concentrations throughout the sessions (n=24)

Analysis of the immediate pre-meal value to five hours later, revealed a main effect of time (F(2, 48.6) = 105.62, p<0.001) that was described primarily by linear (F(1, 23) = 81.29, p<0.001; 31.6% of the variance) and quadratic components (F(1, 23) = 163.23, p <0.001; 63.2% of the variance), with a cubic component that explained less variance (F(1, 23) = 49.09, p<0.001; 4.8% of the variance). There were significant interactions between condition and both the linear (F(1, 23) = 17.86, p<0.001) quadratic components (F(1, 23) = 4.71, p=0.04). Follow-up analyses for the linear component revealed differences such that stair climbing was of smaller magnitude than uninterrupted sitting (p<0.001) and light walking (p=0.02). Similar differences between conditions occurred for the quadratic component but in this case stair climbing was of greater magnitude than uninterrupted sitting (p=0.04) and light walking (p=0.04). There were no differences between the conditions for the cubic component over time (F(1, 23) = 0.24 p=0.63). Inspection of the graph suggests that the NEFA response with stair climbing returned more rapidly towards pre-meal levels after the nadir at two hours.

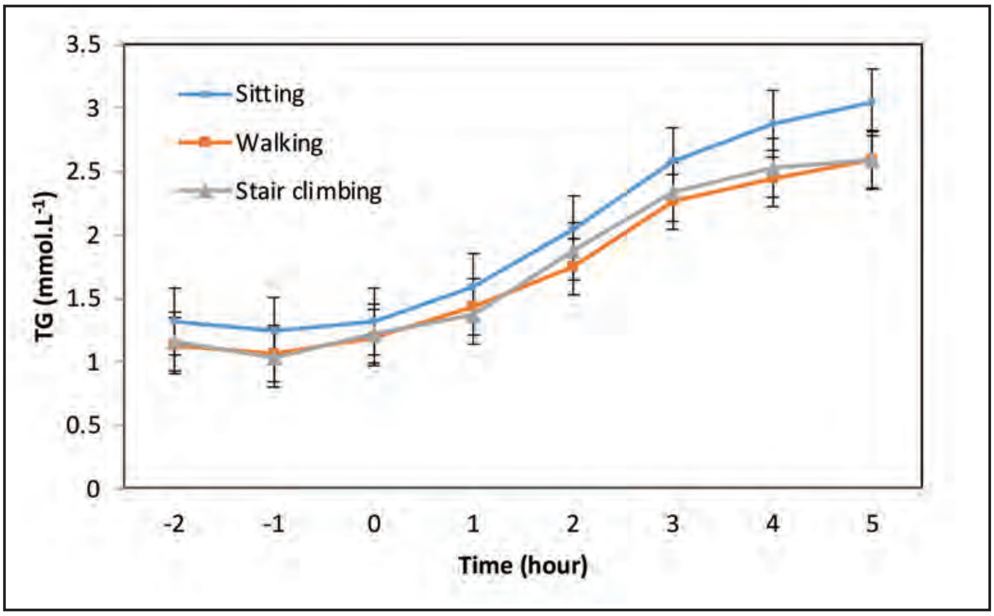

Fig. 2: Triglycerides concentrations throughout the sessions (n=24)

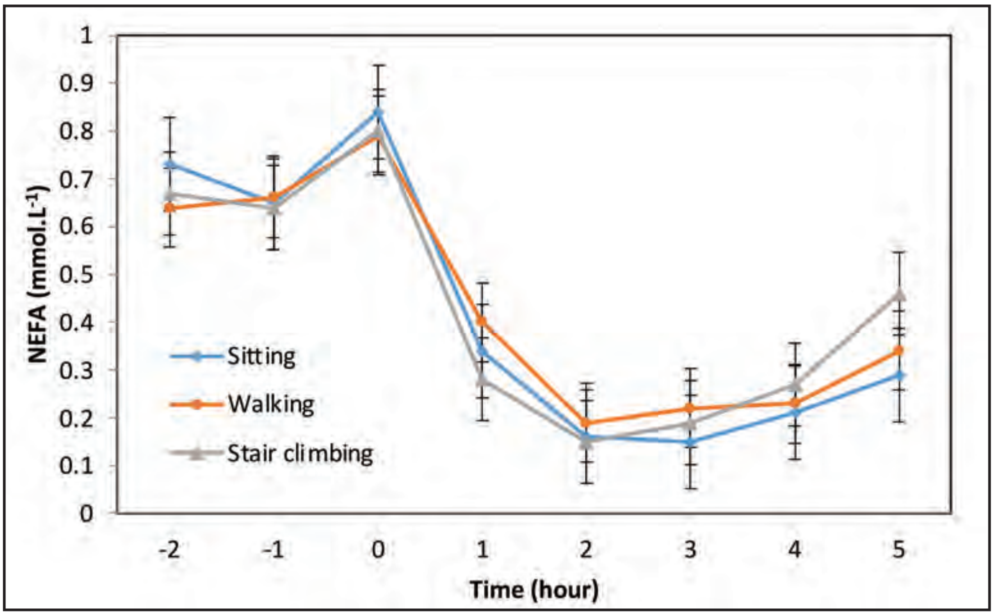

Fig. 3: NEFA concentrations throughout the sessions (n=24)

For the NEFA five-hour AUC, there were no significant differences between conditions (condition linear F(1, 23) =2.86, p=0.104). Similarly, for the two-hour AUC, there was only a marginally significant main effect of condition (F(2, 40.7) = 2.60, p=0.09) suggesting equivalent reductions over two hours postprandially.

DISCUSSION

A mixed pattern was seen in this study. For triglycerides, light intensity walking reduced concentrations postprandially, with only suggestive evidence of a similar effect for the more intense activity of climbing stairs. For glucose, the evidence was reversed. Stair climbing lowered the postprandial peak compared to sitting, with no significant effects of light intensity walking, contrary to Dunstan et al.[5] Nonetheless, stair climbing did not have a significantly greater effects than light walking as predicted. For NEFA, stair climbing resulted in a more rapid return towards pre-meal values than uninterrupted sitting and light walking but unlike Henson et al.,[22] there was no attenuation of NEFA reductions postprandially with light walking. Overall, the data do not confirm the hypothesis of greater effects, the more intense the physical activity used to interrupt sitting.[28]

As outlined in the introduction, light intensity walking has been shown to reduce postprandial glucose in healthy individuals[16,19,20] and in those who were overweight/obese.[5,22]

Mechanically, even light ambulation enhances skeletal muscle glucose uptake through contraction-mediated GLUT4 translocation, independent of insulin which helps dispose of circulating glucose after meals. This effect is particularly relevant postprandially, when skeletal muscle accounts for the majority of glucose disposal. Moreover, breaking prolonged sitting with light walking attenuates the suppression of skeletal muscle lipoprotein lipase (LPL) activity, thereby supporting both lipid and glucose metabolism. In contrast, light intensity walking here had no effect on glucose in a sedentary overweight/obese sample of comparable size to Dunstan and co-workers original study. One possible explanation is that the relative young, lower risk sample here (32 years; fasting glucose 4.29 mmol.L-1) compared to Dunstan and co-workers (54 years; fasting glucose 5.03 mmol.L-1).[5] Younger adults exhibit higher baseline insulin sensitivity and greater cardiorespiratory fitness which can buffer the impact of low-intensity activity. In aging populations by contrast, declines in mitochondrial function, vascular health and insulin sensitivity mean that even light activity provides a proportionally greater benefit for glucose disposal. Typically, cardio-respiratory fitness declines with age as does glucose control. Importantly, BMI and cardio-respiratory fitness exert independent and sometimes opposing effects on glucose homeostasis. McCarthy and co-workers have demonstrated reduced effects of light intensity walking in fitter individuals relative to less fit participants.[19] In contrast, the vigorous activity of stair climbing reduced postprandial glucose, consistent with the effects of walking at moderate intensity in both Peddie et al. and Dunstan et al.[5,13]

As with other metabolic variables, results for the effects of interruptions to sitting on triglycerides postprandially have been mixed. Neither Thorp et al. nor Benatti et al. reported any effects of standing on triglycerides in overweight/obese and healthy samples respectively.[24,25] In contrast, a combination of light walking and standing under free living conditions reduced triglycerides in elderly diabetic individuals the next day.[11] Consistent with the latter study, walking at light intensity and moderate intensity[12] reduced postprandial triglycerides in healthy men and an older sample of healthy post-menopausal women respectively. The reductions in triglycerides with light walking are consistent with two previous studies. What is surprising is that the more intense activity of stair climbing did not have a similar effect. Nonetheless,

Analyses based on orthogonal polynomials were more sensitive to changes than those using the AUC. It is possible that the failure to find consistent effects of the intensity of the interruption reflected a lower risk sample here than in previous research.[5,28] As noted in the methods, changes in biochemical variables have characteristic shapes over time. An overall increase would result in a linear component whereas a peak within the time window would be reflected by a quadratic component. These separable aspects of change over time with polynomials allow independent tests of each aspect of shape in the response. For triglycerides, the consistent increase postprandially was reflected in a substantial linear trend (93% of variance). Both AUC and the linear polynomial analyses concurred that light intensity walking reduced triglycerides postprandially in this data set. In contrast, the glucose response was primarily quadratic in shape (63.6% of the variance). The peak response in the quadratic polynomial was smaller with stair climbing than uninterrupted sitting; there were no AUC differences. For NEFA, statistically similar AUCs contrasted with significant polynomial differences between stair climbing and the other conditions. An AUC provides a single estimate of change over time yet for the glucose polynomials, a substantial cubic component (32.3% of variance) reflected the asymmetrical change as glucose peaked at two hours within a five-hour window. Neither this cubic component nor the minor linear one (2.7% of the variance) differed between conditions. In essence, the glucose AUC included 35% of the variance for glucose change which did not differ between conditions. These two components simply added noise to the glucose AUC. In this study, polynomials were more sensitive to visible differences over time for glucose and NEFA.

CONCLUSION

This study had tested the effects of the intensity of short bouts of physical activity as intermittent interruptions to sitting. Light intensity walking improved triglycerides whereas vigorous intensity stair climbing improved glucose and NEFA when compared to sitting uninterrupted. The results of both AUC and the linear polynomial analyses concurred that light intensity walking reduced triglycerides postprandially in this data set. As it is already known that analyses based on orthogonal polynomials were more sensitive to changes than those using the AUC; hence it is possible that the failure to find consistent effects of the intensity of the interruption reflected a lower risk sample here than those found in previous research.

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