Title: Associations between prolonged sedentary time and breaks in sedentary time with cardiometabolic risk in 10-14 year-old children: the HAPPY study

*Daniel P Bailey[a], Sarah J Charman[b], Thomas Ploetz[c], Louise A Savory[a], Catherine J Kerr[d].

[a]Institute for Sport and Physical Activity Research, School of Sport Science and Physical Activity, University of Bedfordshire, Polhill Avenue, Bedford, United Kingdom.

[b]Institute of Cellular Medicine, Newcastle University, Newcastle upon Tyne, United Kingdom.

[c]Open Lab, School of Computing Science, Newcastle University, Newcastle upon Tyne, United Kingdom.

[d]Allied & Public Health, Faculty of Medical Science, Anglia Ruskin University, East Road Campus, Young Street Site, Cambridge.

***Corresponding author:** Dr. Daniel Bailey, Institute for Sport and Physical Activity Research, School of Sport Science and Physical Activity, University of Bedfordshire, Polhill Avenue, Bedford, Bedfordshire, MK41 9EA, UK. Phone: +441234 793237, email: daniel.bailey@beds.ac.uk.

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Abstract

This study examines the association between prolonged sedentary time and breaks in sedentary time with cardiometabolic risk in 10-14 year-old children. This cross-sectional design study analysed accelerometry-determined sedentary behaviour and physical activity collected over seven days from 111 (66 girls) UK schoolchildren. Objective outcome measures included waist circumference, fasting lipids, fasting glucose, blood pressure, and cardiorespiratory fitness. Logistic regression was used for the main data analysis. After adjustment for confounders, the odds of having hypertriglyceridaemia (p=0.03) and an increased clustered cardiometabolic risk score (p=0.05) were significantly higher in children who engaged in more prolonged sedentary bouts per day. The number of breaks in sedentary time per day was not associated with any cardiometabolic risk factor, but longer mean duration of daily breaks in sedentary time were associated with a lower odds of having abdominal adiposity (p=0.04) and elevated diastolic blood pressure (p=0.01). These associations may be mediated by engagement in light activity. This study provides evidence that avoiding periods of prolonged uninterrupted sedentary time may be important for reducing cardiometabolic disease risk in children.

Introduction

Cardiometabolic risk factors that independently predict cardiometabolic disease in adults include abdominal obesity, hypertension, dyslipidaemia, and impaired fasting glucose (Graham et al., 2007). The clustering of these risk factors confers additive risk beyond the level predicted by the individual component (Golden et al., 2002). This risk factor clustering is increasing in children, with the metabolic syndrome prevalent in 8.6% of American youths (Johnson et al., 2009). This is concerning as risk factor clustering persists into adulthood and increases subsequent risk of cardiometabolic disease (Camhi & Katzmarzyk, 2010; Morrison, Friedman, & Gray-McGuire, 2007). Low cardiorespiratory fitness is also a predictor of increased cardiometabolic risk in children (Bailey, Boddy, Savory, Denton, & Kerr, 2012) and the determinants of these health risk markers thus need to be identified.

Sedentary behaviour is defined as “any waking behaviour characterised by an energy expenditure ≤1.5 METs while in a sitting or reclining position” (Sedentary Behaviour Research Network, 2012). The manner in which sedentary time is accumulated (i.e. engaging in prolonged uninterrupted bouts of sitting) has emerged as a significant cardiometabolic disease risk factor with cross-sectional beneficial associations observed between increased frequency of breaks in sedentary time and cardiometabolic risk in adults (Healy et al., 2008), independent of total sedentary time and moderate-to-vigorous physical activity (MVPA). The few studies examining associations between sedentary bouts and breaks in sedentary time with cardiometabolic risk in children have revealed conflicting findings or have not provided a comprehensive analysis of different sedentary patterns (Altenburg et al., 2015; Carson & Janssen, 2011; Colley et al., 2013; Saunders et al., 2013b). The inconsistent findings may be due to the use of different operational definitions of sedentary bouts and breaks in sedentary time. Several studies tolerated non-sedentary activity within their sedentary bout definitions (Carson & Janssen, 2011; Colley et al., 2013; Saunders et al., 2013b). These permitted interruptions in

sedentary time should not be tolerated as they may attenuate the detrimental effects of prolonged sedentary time on cardiometabolic disease risk. Previous research has also failed to examine the associations between patterns of sedentary time and cardiorespiratory fitness in children.

The primary aims of this study were to examine the association of individual cardiometabolic risk factors, clustered cardiometabolic risk, and cardiorespiratory fitness in 10-14 year-old children with (a) the number of prolonged uninterrupted bouts of sedentary time using an operational definition derived from experimental studies and allowing zero tolerance above the sedentary activity cut-point, (b) duration of prolonged sedentary bouts, (c) number of breaks in sedentary time, and (d) mean duration of daily breaks in sedentary time.

Methodology

Participants

The participants investigated were part of the Health And Physical activity Promotion in Youth (HAPPY) study and the data analysed in this report was collected in 2008. Participants were 10-14 year-old children recruited on a voluntary basis in 11 schools across Bedfordshire, UK, and baseline data from 45% of the total sample was analysed in the present study. Participants were excluded from the HAPPY study if they were on medication for high blood pressure or if they had heart conditions, dizziness, or joint pain that could be exacerbated through exercise. As part of the consent process, parents were given the option to remove their child from the blood sampling procedure. Further participants were excluded if they did not reach the inclusion criteria specified below for physical activity or cardiorespiratory fitness assessment. Participants who consented to providing blood samples engaged in significantly more light activity and MVPA than those who did not consent (p=0.02 and 0.05, respectively), while there was no difference in sedentary behaviour variables (p>0.05). The HAPPY study received full ethical approval from the University of Bedfordshire Ethics Review Board. Written informed consent was obtained from participants' parents and verbal assent from the participants before any test procedures.

Measures

Participants were required to fast from 9 pm the night before measurements were performed. Body composition and blood measurements were taken between 8-10 am and participants were instructed to bring a snack with them to eat for breakfast afterwards. Stature and waist circumference (WC) measured at the umbilicus were recorded to the nearest 0.5 cm using the portable Leicester Height Measure (Seca, Birmingham, UK) and an adjustable tape measure (Hoechstmass, Sulzbach, Germany), respectively. Body mass and body fat% were recorded to the nearest 0.1

kg and 0.1%, respectively, using the Tanita BC-418 Segmental Body Composition Analyzer (Tanita Corp., Tokyo, Japan), which is valid for epidemiological studies assessing whole body fat in children (Luque et al., 2014). Body mass index (BMI) was calculated using the equation: BMI = body mass (kg) ÷ stature^2 (m^2). UK reference values were used to calculate *z*-scores for height, weight, BMI, and WC (*z*-WC) (Cole, Freeman, & Preece, 1995; Freeman et al., 1995; McCarthy, Jarrett, & Crawley, 2001).

Sitting blood pressure was measured using an Omron M5-I automated oscillatory device (Omron Matsusaka Co. Ltd., Matsusaka, Japan) on the left arm after the participant had rested for 5 min. Three blood pressure readings were obtained with 2 min rest between each and the average of the lowest two readings recorded. Fasting blood samples (40 µl) were obtained using a finger prick method and analysed using a point-of-care Cholestech LDX Analyzer (Cholestech Corp., Hayward, CA.) to determine total cholesterol (TC), high-density lipoprotein cholesterol (HDL), triglycerides, and blood glucose levels. This system is validated in adults (Parikh, Mochari, & Mosca, 2009) and is certified by the Centers for Disease Control Cholesterol Reference Method Laboratory Network, This system has been used in previous paediatric research (Ahrens et al., 2014).

Cardiorespiratory fitness was determined indirectly using a previously validated age- and sex-specific all-out progressive cycle ergometer test to exhaustion (Riddoch et al., 2005) and this took place a minimum of 90 min after the breakfast snack. Workloads increased every 3 min until volitional exhaustion occurred. A maximal effort was deemed as a final heart rate ≥185 bpm in addition to subjective observation from the researcher that the participant could not continue. Participants who did not achieve a maximal effort were excluded from the analysis. Power output (watts) was calculated as being equal to $W_1 + (W_2 \cdot t/180)$, where W_1 is work rate at fully completed stage, W_2 is the work rate increment at final incomplete stage, and *t* is time in seconds at the final incomplete stage. $\dot{V}O_{2max}$ was

estimated using previously described formulae (Hansen, Froberg, Nielsen, & Hyldebrandt, 1989) and expressed as mL per kilogram of body mass per min ($mL^{-1}/kg^{-1}/min^{-1}$). Estimated $\dot{V}O_{2max}$ using this approach correlates significantly with directly assessed $\dot{V}O_{2max}$ in boys and girls (r=0.90 and 0.95, respectively, p<0.01) with a standard error of estimation of 3.2% (Hansen et al., 1989).

Participants were then asked to wear a tri-axial accelerometer (RT3®, Stayhealthy, Inc., Monrovia, CA) on their dominant hip to measure sedentary behaviour and physical activity over the next seven days during waking hours in 1-min epochs. Sequences of ≥10 min of consecutive zero counts were removed during the recoding process (Riddoch et al., 2004). The inclusion criteria for analysis were a minimum wear time of three days (Mattocks et al., 2008) and a minimum daily wear time of nine hours for weekdays (Mattocks et al., 2008) and eight hours for weekend days (Rowlands, Pilgrim, & Eston, 2008). Participants not meeting these criteria were excluded from the present analysis. There is currently substantial variation in cut-points used to define sedentary behaviour and physical activity intensities (Reilly et al., 2008). For this analysis, time spent sedentary (<420 counts per minute [cpm]), in light physical activity (420-1859 cpm), and in MVPA (≥1860 cpm) was determined from a previous validation study in children (Chu, 2007). The sedentary cut-point was determined solely from non-ambulation activities (lying, sitting, and static standing) and the threshold of 420 cpm provided a sensitivity and specificity of 100% for distinguishing sedentary behaviour from low-intensity ambulation (Chu, 2007). The total number of prolonged sedentary bouts (≥20 min of uninterrupted sedentary time), mean duration of prolonged sedentary bouts, number of breaks in sedentary time (defined as a sedentary bout interrupted by ≥1 min of light activity or MVPA), and mean duration of daily breaks in sedentary time were calculated. All physical activity and sedentary variables were calculated each day and then averaged across valid days. The 20 min cut-point to define a prolonged

sedentary bout was based on experimental research in children and young adults demonstrating deleterious cardiometabolic effects of engaging in ≥20 min uninterrupted prolonged sitting bouts (Bailey & Locke, 2015; Belcher et al., 2015). The 1 min cut-point to define a break in sedentary time was based on evidence demonstrating beneficial associations with cardiometabolic risk in adults (Healy et al., 2008). Similar to previous research, participants were stratified into groups (low and high) based on the sample mean for each individual sedentary behaviour variable (Carson & Janssen, 2011).

Abdominal obesity was defined as WC ≥90th percentile for age and sex (McCarthy et al., 2001). Hypercholesterolaemia was defined as TC ≥5.17 mmol/L (National Cholesterol Education Panel, 1991). The National Cholesterol Education Program's Pediatric Panel Report (1991) gives a range of 0.91-1.16 mmol/L for borderline low HDL levels and 1.02-1.46 mmol/L for borderline high triglyceride concentrations for all sexes and ages. Therefore, the midpoint of these ranges was used as the 10th percentile value to define low HDL (≤1.03 mmol/L) and the 90th percentile value to define hypertriglyceridaemia (≥1.24 mmol/L) (Cook, Weitzman, Auinger, Nguyen, & Dietz, 2003). Impaired fasting glucose was defined as ≥5.6 mmol/L (Zimmet et al., 2007). High systolic and diastolic blood pressure was defined as ≥90th percentile for age, sex, and height (National High Blood Pressure Education Program Working Group on High Blood Pressure in Children and Adolescents, 2004). Metabolic syndrome was defined as having ≥3 of the following cardiometabolic risk factors: abdominal obesity (high WC), low HDL, hypertriglyceridaemia, high systolic or diastolic blood pressure, and impaired fasting glucose. Cardiorespiratory fitness values >37.0 $mL^{-1}/kg^{-1}/min^{-1}$ for girls and >42.1 $mL^{-1}/kg^{-1}/min^{-1}$ for boys represented a high cardiorespiratory fitness level, while values below these levels represented low cardiorespiratory fitness (Ruiz et al., 2007).

A continuous clustered cardiometabolic risk score was also calculated as this approach increases statistical power (Ragland, 1992) and is used in paediatric research (Saunders et al., 2013b). First, TC:HDL ratio and triglycerides were non-normally distributed and were log-transformed. The clustered risk score was then constructed by summing the standardised *z*-scores for the following continuously distributed variables: WC, diastolic blood pressure, TC:HDL ratio, triglycerides, and blood glucose. Participants were then assigned to a 'normal' or 'at-risk' clustered cardiometabolic risk group with high risk defined as ≥1 SD (2.77) in risk score above the pooled mean (Andersen et al., 2006). A non-obesity clustered risk score was also calculated by removing *z*-WC (Ekelund et al., 2007). High risk for the non-obesity clustered risk score was defined as ≥1 SD (2.28) in risk score above the pooled mean.

Potential covariates included age, sex, ethnicity (recorded as white or non-white), and socioeconomic status, which was determined using the 2007 Indices of Multiple Deprivation (Department for Communities and Local Government, 2008).

Statistical analysis

Analyses were completed using IBM SPSS Statistics version 21.0 (SPSS Inc., Armonk, N.Y., USA). Descriptive data is expressed as mean±SD. Partial correlation analysis explored relationships between sedentary behaviour variables and cardiometabolic risk factors. Of the potential covariates, sex and ethnicity were significantly related with ≥1 cardiometabolic risk factor and were thus adjusted for in the analysis in addition to total sedentary time (except where total sedentary time was an independent variable), MVPA, and accelerometer wear time (Altenburg et al., 2015). Abdominal obesity (*z*-WC) was additionally adjusted for in the analysis exploring relationships with cardiorespiratory fitness and the non-obesity clustered cardiometabolic risk score.

196 Multiple logistic regression models were used to examine the odds of having
197 individual and clustered cardiometabolic risk factors according to high and low
198 levels for the sedentary bout and breaks in sedentary time variables. Participants
199 categorised into the low group were the reference comparator for each independent
200 sedentary behaviour variable and dependent cardiometabolic risk factors, except for
201 HDL where participants with high levels were the reference comparator. All
202 regression models were adjusted for the same variables as described above for the
203 correlation analysis (model 1). Light activity may be independently associated with
204 cardiometabolic risk (Healy et al., 2007) and all regression models were therefore
205 performed again additionally adjusting for this variable to explore whether light
206 activity mediates the associations between sedentary behaviour variables and
207 cardiometabolic risk (model 2). Data for the regression analysis is expressed as
208 odds ratio (OR) and 95% confidence interval (CI). The level of significance was set
209 at $p \leq 0.05$.
210

Results

Of the 249 participants from the HAPPY study, 93 did not provide consent to blood sampling and were excluded from the present analysis. Of the remaining 156 participants, 45 participants did not provide valid accelerometry data and were excluded from the present analysis. This resulted in 111 participants (66 girls, 11.8±1.4 years) being included in the final analysis. Three participants did not achieve a maximal effort during the cardiorespiratory fitness test and associations between sedentary behaviour variables and cardiorespiratory fitness are thus reported for 108 participants.

Table 1 shows the descriptive characteristics of the participants. The prevalence of abdominal adiposity was 17.1%; hypercholesterolaemia 6.3%; low HDL 12.6%; hypertriglyceridaemia 17.1%; elevated systolic blood pressure 18.9%; elevated diastolic blood pressure 18.0%; and impaired fasting glucose 11.7%. Metabolic syndrome was prevalent in 7.2% of participants, 16.2% had an increased clustered cardiometabolic risk score, and 18.9% had an increased non-obesity clustered cardiometabolic risk score. The prevalence of low cardiorespiratory fitness was 35.2%.

After adjusting for sex, ethnicity, total sedentary time, MVPA, and accelerometer wear time, the duration of prolonged sedentary bouts was significantly positively correlated with systolic blood pressure (Table 2). The number of breaks in sedentary time was significantly negatively correlated with cardiorespiratory fitness. The mean duration of daily breaks in sedentary time was significantly positively correlated with cardiorespiratory fitness and significantly negatively correlated with total cholesterol. Total sedentary time was significantly negatively correlated with abdominal adiposity.

The multivariate adjusted ORs (and 95% CIs) for having adverse cardiometabolic risk factor levels across low and high groups for the number of prolonged sedentary bouts (5.5±1.2 and 8.4±0.8 for the low and high groups, respectively), duration of prolonged sedentary bouts (33.25±2.36 and 40.90±4.17 min/day for the low and high groups, respectively), number of breaks in sedentary time (53.0±6.4 and 72.6±10.4 for the low and high groups, respectively), and mean duration of daily breaks in sedentary time (3.15±0.38 and 4.46±0.63 min/day for the low and high groups, respectively) are presented in Table 3. After adjusting for sex, ethnicity, total sedentary time, MVPA, and accelerometer wear time (model 1), the odds of having hypertriglyceridaemia, increased clustered cardiometabolic risk score, and increased non-obesity clustered cardiometabolic risk score were significantly higher in children who engaged in more prolonged sedentary bouts per day. The duration of prolonged sedentary bouts and the number of breaks in sedentary time were not significantly associated with cardiometabolic risk. Children who engaged in longer mean duration daily breaks in sedentary time had significantly lower odds of having abdominal obesity and elevated diastolic blood pressure. When additionally adjusting for light activity (model 2), the associations between the number of prolonged sedentary bouts with hypertriglyceridaemia (OR=3.91, 95% CI 0.79, 19.29, p=0.10), clustered cardiometabolic risk score (4.31; 0.79, 23.60, p=0.09), and the non-obesity clustered cardiometabolic risk score (4.19; 0.87, 20.29, p=0.08) were attenuated and no longer significant. The higher odds of having abdominal obesity in children who engaged in longer duration prolonged sedentary bouts strengthened and became significant (5.00; 0.98, 25.43, p=0.05). The lower odds of having abdominal obesity (0.10; 0.01, 0.80, p=0.03) and elevated diastolic blood pressure (0.09; 0.01, 0.68, p=0.02) in children who engaged in longer mean duration daily breaks in sedentary time was unaffected.

Discussion

The main findings of this study were that engaging in more prolonged sedentary bouts per day was significantly associated with increased cardiometabolic risk in 10-14 year-old children, while engaging in longer mean duration daily breaks in sedentary time was significantly associated with lower cardiometabolic risk .

The children in this study engaged in an average of seven prolonged sedentary bouts (≥20 min) per day lasting an average 37 min. In other studies applying the strictest definition of uninterrupted sedentary time (i.e. zero tolerance), Saunders et al. (2013b) found that 8-11 year-olds engaged in five sedentary bouts per day lasting 15-29 min, whereas Altenburg et al. (2015) reported 10-13 year-old children engaged in an average of two sedentary bouts lasting ≥20 min per day. However, Altenburg et al. (2015) used a different accelerometer model to the present study and a shorter epoch length for data capture (15-s versus 1-min in the present study), which could explain the discrepancies observed.

Engaging in more prolonged sedentary bouts was associated with higher odds of hypertriglyceridaemia and increased clustered cardiometabolic risk (independent of abdominal obesity) in the present study. It may thus be appropriate for paediatric health promotion strategies to target reductions in the number of prolonged sedentary bouts accumulated per day to reduce cardiometabolic disease risk. In 6-19 year-old children, no associations between sedentary time accumulated in ≥20 min and ≥30 min bouts and cardiometabolic risk were observed (Carson & Janssen, 2011; Colley et al., 2013). This was in addition to ≥30, 60, 100, and 120 min sedentary bout durations, although positive associations between bouts lasting ≥40 and 80 min with anthropometrics were observed in a subgroup of 10-14 year-old boys (Colley et al., 2013). Each of these above studies permitted non-sedentary activity within their sedentary bout definitions, which may explain the lack of associations reported. When allowing zero tolerance time within sedentary bouts, Altenburg et al. (2015) did not observe associations between ≥20 min

sedentary bouts and cardiometabolic risk, but did report weak associations with sedentary time accumulated in bouts of ≥5, 10, and 30 min. The reason prolonged bouts of ≥20 min was not associated with cardiometabolic risk is not clear but may be due to the low number of ≥20 min sedentary bouts observed (n=2), thus providing insufficient statistical power (Altenburg et al., 2015). Saunders et al. (2013b) allowed zero tolerance time in their sedentary bout definition and reported a number of negative and positive associations of sedentary bout durations lasting 1-4 min, 5-9 min, 10-14 min, 15-29 min, and ≥30 min with various cardiometabolic risk factors. However, using this delimited approach makes the findings difficult to interpret and compare to the present study. Future studies should thus use consistent definitions of sedentary time bouts and patterns. Based on previous experimental research in children (Belcher et al., 2015) and the findings of the present study, we suggest defining a prolonged sedentary bout as ≥20 min of uninterrupted sedentary time with zero tolerance of non-sedentary activity within bouts.

Adjusting for light activity attenuated the associations between the number of prolonged sedentary bouts with hypertriglyceridaemia and clustered cardiometabolic risk. It is unknown whether light activity would have mediated associations between sedentary bouts and breaks in sedentary time in previous studies as no adjustment for this variable was made (Altenburg et al., 2015; Carson & Janssen, 2011; Colley et al., 2013; Saunders et al., 2013b). As light activity may be an important mediating factor, future studies should account for this variable to identify true independent associations between sedentary behaviour patterns and cardiometabolic risk.

Children in this study engaged in an average of 63 breaks in sedentary time per day. In 6-19 year-olds, the number of daily breaks in sedentary time was 81 and 85 in boys and girls, respectively (Colley et al., 2013), Carson, Stone, and Faulkner (2014) reported that 11 year-old children engaged in 56 breaks per hour, which is substantially higher than that reported previously and in the present study. However,

Carson et al. (2014) defined a break in sedentary time as any 5-s epoch change above the sedentary activity cut-point, whereas a break was defined as an interruption lasting ≥1 min in the present study and previous research (Colley et al., 2013). The duration of breaks in sedentary time has not been reported in previous paediatric research. The mean duration of daily breaks in sedentary time in the present study was 3.8 min, which is comparable to adults (Healy et al., 2008). Definitions of a break in sedentary time varies widely (Chastin, Egerton, Leask, & Stamatakis, 2015) and it is thus difficult to propose a standardised definition for this variable. Further research should establish the minimum duration of a break in sedentary time required to produce cardiometabolic benefits.

The number of breaks in sedentary time was not associated with cardiometabolic risk or cardiorespiratory fitness in the present study, which supports previous findings (Carson & Janssen, 2011). However, in 8-11 year-old children, the number of breaks in sedentary time was negatively associated with clustered cardiometabolic risk (Saunders et al., 2013b). The reason for this disparity is not clear as both the present study and Saunders et al. (2013b) used 1-min accelerometer epochs and applied the same definition of a break in sedentary time. Saunders et al. (2013b) studied children with a family history of obesity who were slightly younger than the current sample. The present study used a triaxial accelerometer, whereas Saunders et al. (2013b) used a uniaxial model that only captures vertical accelerations and these could be explanatory factors. The present study is the first to examine associations between the mean duration of daily breaks in sedentary time and cardiometabolic risk in children. Longer mean duration daily breaks in sedentary time were associated with lower odds of abdominal obesity and elevated diastolic blood pressure. This suggests that in addition to reducing the number of prolonged sedentary bouts, children should also increase the duration of breaks in sedentary time to reduce cardiometabolic disease risk.

The present study is the first to examine associations between prolonged sedentary time and cardiorespiratory fitness in children. When participants were stratified into low and high groups for each of the sedentary behaviour variables, there were no associations with cardiorespiratory fitness. However, the number of breaks in sedentary time was negatively correlated with cardiorespiratory fitness, while the mean duration of daily breaks in sedentary time was positively correlated. The greater volume of physical activity accumulated through longer mean duration daily breaks in sedentary time may be sufficient to promote a higher level of cardiorespiratory fitness. The reason more breaks in sedentary time was correlated with lower cardiorespiratory fitness is not clear and is suggestive of a complex interaction between this health risk marker and sedentary behaviour patterns. Further research is needed to establish the importance of sedentary bouts and breaks in sedentary time for cardiorespiratory fitness in children.

There are several potential mechanisms that could explain the detrimental association of prolonged sedentary bouts with cardiometabolic risk. In adults, imposed prolonged sedentary behaviour can cause an acute reduction in insulin action, lower HDL, and increase glucose and triglyceride concentrations (Saunders, Larouche, Colley, & Tremblay, 2012). These deleterious effects may be partially mediated by reductions in lipoprotein lipase activity, which facilitates uptake of free fatty acids into muscle and adipose tissue, and increases in glucose uptake stimulated by insulin and the glucose transporter protein GLUT-4 (Hamilton, Hamilton, & Zderic, 2007). Imposed sedentary behaviour causes an acute decline in vascular function in adults and young females, which appears to be mediated by reductions in shear stress that influences the structure of blood vessels and endothelial cell function (McManus et al., 2015; Thosar, Bielko, Mather, Johnston, & Wallace, 2015). In adults, the acute declines in cardiometabolic function can be mitigated by breaking up prolonged sedentary time with regular short bouts of physical activity (Chastin et al., 2015), whereas in children the evidence is not

consistent (Belcher et al., 2015; Heden, Liu, Park, Winn, & Kanaley, 2015; McManus et al., 2015; Ross, Hinckson, & Zinn, 2015; Saunders et al., 2013a). With regards to abdominal obesity, it is plausible that children who engage in longer duration breaks in sedentary time expend more daily energy and therefore store less energy as fat.

The strengths of this study include objectively measured sedentary behaviour patterns and cardiometabolic risk factors and detailed analysis of sedentary time using strict definitions. Limitations of this study include the cross-sectional design, thus limiting conclusions regarding causality. There is a lack of consensus regarding the most appropriate cut-off points for metabolic syndrome risk factors in children and this could affect the associations observed. As a waist-worn accelerometer was used, it is possible that some light activities (e.g. standing still) were misclassified as sedentary behaviour, which may have led to an overestimation of sedentary time. The use of 1-min epochs and potential measurement error of the accelerometer may have led to misclassification of sedentary time or physical activity, which could have affected the associations observed and the appropriateness of the standardised recommendations for sedentary bouts and breaks in sedentary time. Future studies should consider the instrument and epoch duration to be used so that definitive conclusions regarding the association of sedentary behaviour patterns with cardiometabolic risk in children can be made. A minimum of three days accelerometer wear time was required in the present analysis and periods of ≥10 min of consecutive zero accelerometer counts were defined as non-wear time. A minimum of three days is often used in the literature (Toftager et al., 2013) but the minimum number of days to produce reliable data using the RT3® accelerometer is unknown. Previous research has applied criteria of between 10 and 60 min of consecutive zero counts (Toftager et al., 2013). The most appropriate criteria to classify non-wear time is unknown and a consensus is needed to provide greater comparability between studies. Adjusting for light

activity attenuated some of the associations observed between the number of prolonged sedentary bouts and cardiometabolic risk markers. Although these associations became non-significant, it is possible that significance would have remained with a larger sample size. There was no measure of maturation available in this study, which could have confounded the associations observed as transient changes in cardiometabolic risk occur during puberty (Moran et al., 2008). The potential confounding effects of dietary intake and eating behaviour that are independently associated with cardiometabolic risk (Eloranta et al., 2014) were also not accounted for. Other potential confounding factors not accounted for include smoking and the presence of Type 1 or Type 2 diabetes.

In conclusion, the findings of the present study demonstrate that prolonged sedentary bouts and the mean duration of daily breaks in sedentary time are independently associated with cardiometabolic risk in 10-14 year-old children. Future research should aim to use standardised methods of measurement and definitions of sedentary bouts and breaks in sedentary time. These standardised approaches should then be utilised in longitudinal and experimental studies to determine which patterns of sedentary time are important for paediatric health.

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591

592 **Table 1** Descriptives for 10-14 year-old UK children

	Total (*n*=111)
Age (years)	11.8±1.4
Gender (%)	
Male	34
Female	66
Ethnicity (%)	
White	78
Non-white	22
z-height	0.36±1.01
z-weight	0.11±1.14
Body fat%	20.7±6.6
Body mass index *z*-score	-0.18±1.33
Cardiometabolic risk markers	
Waist circumference *z*-score	0.16±1.50
Systolic blood pressure (mm Hg)	108.7±9.7
Diastolic blood pressure (mm Hg)	68.6±6.9
Total cholesterol (mmol/L)	3.93±0.69
HDL (mmol/L)	1.43±0.39
TC:HDL ratio	2.95±1.07
Triglycerides (mmol/L)	0.85±0.49
Blood glucose (mmol/L)	5.04±0.46
Cardiorespiratory fitness ($mL^{-1}/kg^{-1}/min^{-1}$)	42.06±9.54
Accelerometer-derived variables	
Number of prolonged sedentary bouts per day	7.0±1.8
Duration of prolonged sedentary bouts (min/day)	37.11±5.12
Number of breaks in sedentary time per day	62.9±13.0
Mean duration of daily breaks in sedentary time (min/day)	3.80±0.83
Total sedentary time (min/day)	504.02±80.53
Light physical activity (min/day)	182.44±47.01
Moderate-to-vigorous physical activity (min/day)	45.04±26.35
Total wear time (min/day)	731.84±72.87

593 Data presented as mean±SD.

594 HDL, high-density lipoprotein cholesterol; TC, total cholesterol.

595 *n*=108 for cardiorespiratory fitness.

596

Table 2 Partial correlations between sedentary time and patterns and cardiometabolic risk factors in 10-14 year-old children

	Total sedentary time (min/day)	Number of prolonged sedentary bouts per day	Duration of prolonged sedentary bouts (min/day)	Number of breaks in sedentary time per day	Mean duration of daily breaks in sedentary time (min/day)
z-WC	**-0.23**	-0.02	0.11	0.05	-0.09
Total cholesterol (mmol/L)	0.02	-0.09	-0.09	0.09	**-0.24**
HDL (mmol/L)	0.02	-0.12	0.02	-0.03	0.07
Triglycerides (mmol/L)[a]	-0.16	0.14	0.04	-0.18	0.09
Blood glucose (mmol/L)	0.12	0.07	0.08	-0.12	0.08
Systolic blood pressure (mmHg)	-0.09	0.05	**0.21**	-0.06	-0.03
Diastolic blood pressure (mmHg)	-0.16	0.05	0.13	0.03	-0.07
Clustered cardiometabolic risk score	-0.12	0.12	0.12	-0.06	-0.08
Non-obesity clustered cardiometabolic risk score	-0.01	0.16	0.02	-0.11	-0.05
Cardiorespiratory fitness ($mL^{-1}/kg^{-1}/min^{-1}$)	0.14	0.12	0.10	**-0.24**	**0.27**

z-WC, waist circumference *z*-score; HDL, high-density lipoprotein cholesterol.

[a]log transformed.

Partial correlations adjusted for sex, ethnicity, total sedentary time, moderate-to-vigorous physical activity, and accelerometer wear time.

Significant associations ($p \leq 0.05$) highlighted in bold.

Table 3 Multivariate-adjusted odds ratios for cardiometabolic risk factors across sedentary behaviour pattern groups for 10-14 year-old children

	Number of prolonged sedentary bouts per day		Duration of prolonged sedentary bouts (min/day)		Number of breaks in sedentary time per day		Mean duration of daily breaks in sedentary time (min/day)	
	Odds ratio (95% CI)	*P* value	Odds ratio (95% CI)	*P* value	Odds ratio (95% CI)	*P* value	Odds ratio (95% CI)	*P* value
Abdominal obesity[1]	2.39 (0.51, 11.21)	0.27	4.07 (0.92, 18.11)	0.07	1.34 (0.26, 6.80)	0.73	**0.12 (0.02, 0.87)**	**0.04**
Hypercholesterolaemia[2]	0.26 (0.02, 2.8)	0.27	0.98 (0.12, 8.11)	0.99	0.33 (0.02, 5.17)	0.43	1.06 (0.06, 17.88)	0.97
Low HDL[3]	2.30 (0.47, 11.21)	0.30	0.58 (0.15, 2.21)	0.43	3.11 (0.47, 20.59)	0.24	1.51 (0.27, 8.50)	0.64
Hypertriglyceridaemia[4]	**5.25 (1.18, 23.45)**	**0.03**	0.92 (0.26, 3.25)	0.89	1.64 (0.35, 7.75)	0.53	2.11 (0.45, 10.01)	0.35
Impaired fasting glucose[5]	0.20 (0.03, 1.13)	0.07	1.09 (0.27, 4.43)	0.90	0.46 (0.07, 2.99)	0.41	2.71 (0.43, 16.96)	0.29
Elevated systolic BP[6]	0.84 (0.21, 3.32)	0.80	1.55 (0.46, 5.24)	0.48	1.27 (0.29, 5.56)	0.75	0.35 (0.07, 1.86)	0.22
Elevated diastolic BP[6]	1.01 (0.23, 4.54)	0.99	1.38 (0.36, 5.35)	0.64	0.91 (0.18, 4.54)	0.91	**0.07 (0.01, 0.53)**	**0.01**
Metabolic syndrome[7]	3.78 (0.47, 30.40)	0.21	0.50 (0.08, 3.27)	0.47	0.57 (0.06, 5.50)	0.63	0.62 (0.06, 6.15)	0.69
Increased clustered cardiometabolic risk[8]	**4.99 (0.99, 25.10)**	**0.05**	1.08 (0.29, 4.09)	0.91	1.10 (0.21, 5.68)	0.91	0.49 (0.09, 2.66)	0.49
Increased non-obesity clustered cardiometabolic risk[9]	**6.02 (1.29, 28.12)**	**0.02**	0.80 (0.23, 2.86)	0.74	1.25 (0.27, 5.86)	0.78	1.70 (0.35, 8.28)	0.51
High cardiorespiratory fitness[10]	1.82 (0.49, 6.74)	0.37	0.71 (0.20, 2.53)	0.60	2.09 (0.47, 9.35)	0.34	2.54 (0.58, 11.11)	0.22

CI, confidence interval; HDL, high-density lipoprotein cholesterol; BP, blood pressure.

Participants categorised into the low group were the reference comparator for each independent sedentary behaviour variable and dependent cardiometabolic risk factor.

n=55 in the low groups for number of prolonged sedentary bouts per day, time in prolonged sedentary bouts, and number of breaks in sedentary time per day; *n*=56 in the low group for duration of breaks in sedentary time.

Model 1: adjusted for sex, ethnicity, total sedentary time, moderate-to-vigorous physical activity, and accelerometer wear time.

Model 2: adjusted for covariates in Model 1 and light physical activity.

Significant associations ($p \leq 0.05$) highlighted in bold.

[1]≥90th percentile for age and sex; [2]≥5.17 mmol/L; [32] ≤1.03; [4]≥1.24 mmol/L; [5]≥5.6 mmol/L; [6]≥90th percentile for age, sex and height; [7]≥3 of the following risk factors: abdominal obesity, low HDL, hypertriglyceridemia, high systolic or diastolic BP, and impaired fasting glucose; [8]clustered risk score ≥2.77; [9]non-obesity clustered risk score ≥2.28; [10]>42.1 and 37.0 $mL^{-1}/kg^{-1}/min^{-1}$ for boys and girls, respectively.