Title: Associations of objectively measured sedentary behaviour and physical activity with markers of cardio-metabolic health

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Abstract

Aims/hypothesis To examine the associations between objectively measured sedentary time, breaks in sedentary time, moderate-to-vigorous physical activity (MVPA) and total physical activity with markers of cardio-metabolic health in a population with known risk factors for type 2 diabetes mellitus.

Methods This study reports data from two ongoing diabetes prevention programmes. Participants with known risk factors were recruited from primary care practices located within the East Midlands, United Kingdom, 2010-2011. Actigraph GT3X accelerometers (15s epochs) were used to assess sedentary time (<25counts per 15s), MVPA (≥488counts per 15s) and total physical activity (total counts). A break was considered as any interruption in sedentary time (≥25counts per 15s). Linear regression examined the independent association of sedentary time, breaks in sedentary time, MVPA and total physical activity with markers of cardio-metabolic health.

Results The sample comprised 878 participants; 153 from 'Project STAND' (age=32.9±5.6years; female=71.2%) and 725 from 'Walking Away from Diabetes' (age=63.7±7.8years; female=35.2%). Following adjustment for various covariates, including MVPA and BMI, there were detrimental linear associations of sedentary time with 2-hour plasma glucose (standardised beta co-efficient (β)) (β=0.220, p=<0.001), triacylglycerol (β=0.206, p=0.001) and HDL cholesterol (β=-0.123, p=0.029). Breaks in sedentary time, total physical activity and MVPA were significantly inversely associated with measures of adiposity, but not with any other cardio-metabolic variables after adjustment for sedentary time and BMI.

Conclusions/Interpretation In adults at high risk of type 2 diabetes mellitus, time spent sedentary is strongly and adversely associated with cardio-metabolic health and may be a more important indicator of poor health than MVPA.

Walking Away from Type 2 Diabetes Study - **ISRCTN31392913**.
Project STAND (**S**edentary **T**ime **An**d **D**iabetes) - **ISRCTN08434554**.

Keywords: Breaks in sedentary time, High-risk, Physical activity, Primary care, Sedentary behaviour, Type 2 diabetes mellitus.

Abbreviations:
β- Beta-coefficient
cpm- Counts per minute
CVD- Cardiovascular disease
FPG- Fasting plasma glucose
IFG- Impaired fasting glycaemia
IGR- Impaired glucose regulation
IGT- Impaired glucose tolerance
IMD- Index of multiple deprivation
MVPA- Moderate-to-vigorous physical activity
METs- Metabolic equivalents
STAND- **S**edentary **T**ime **An**d **D**iabetes
UK- United Kingdom
WA- Walking Away from type 2 diabetes

Introduction

Sedentary behaviour has previously been characterised as ≤1.5 metabolic equivalents (METs) [1, 2]. METs are the energy cost of physical activity and are expressed as multiples of resting metabolic rate, where one MET (or 3.5 ml min^{-1} kg^{-1}) is equivalent to typical metabolism at rest for an average person. Given the fact it is impractical to measure energy expenditure in most studies and there are limited behaviours that involve both sitting and energy expenditure (>1.5 METs), a more operational behavioural interpretation has been recommended whereby sedentary behaviour is defined as any non-exercise sitting time [3]. Over the last decade, sedentary behaviour has emerged as a distinctive behavioural paradigm with detrimental effects on chronic disease risk, independent of moderate-to-vigorous intensity physical activity (MVPA) [4-8]. This new paradigm is conceptualised around two constructs: total time spent sedentary and the number of breaks in sedentary time (e.g. rising from a sitting/lying position to a more active state, including standing). Both expressions show strong associations with markers of cardio-metabolic health independent of each other and other lifestyle behaviours [4-6, 8, 9].

Epidemiological evidence examining the effect of time spent sedentary has tended to focus on self-report measures [6, 10-12]; these are prone to bias and have poor levels of validity [13]. Although several studies employing objective measures of sedentary behaviour have been reported, the effect of age on the association between sedentary time and cardio-metabolic risk remains unclear and most have been conducted in the general population without reference to specific risk factors [4, 5, 8, 14, 15]. It is therefore unclear to what extent the reported associations are generalisable to those at high risk of chronic disease. This is an important limitation as international recommendations and policies specify that chronic disease prevention strategies should include targeted interventions aimed at the identification and management of high risk individuals [16-18]. Therefore, the importance of sedentary behaviour in this group needs to be better understood in order to inform the content and structure of prevention programmes.

In this study, we examined the extent to which sedentary time, breaks in sedentary time, MVPA and total physical activity are independently associated with cardio-metabolic risk factors in a population with known risk factors for type 2 diabetes mellitus. We hypothesised that all four constructs would be independently associated with health in both younger and older adults.

Methods

Participants This study used combined baseline data from two prevention studies, Walking Away from Type 2 Diabetes Study (WA) (ISRCTN31392913) and Project STAND (**S**edentary **T**ime **An**d **D**iabetes) (ISRCTN08434554), 2010-2011. Both trial protocols have been published elsewhere [19, 20]. Briefly, WA is a randomised controlled trial investigating whether a lifestyle intervention programme can promote behaviour change in those identified at high risk of type 2 diabetes mellitus. Similarly, Project STAND is a randomised controlled trial investigating the effect of structured education and self monitoring on reducing sedentary time in young adults with known risk factors for type 2 diabetes mellitus.

Individuals were unaware of their diabetes risk status before entering both studies and all participants were excluded if they had known type 2 diabetes mellitus or were taking steroids. Baseline measurements across both studies were performed before treatment allocation by the

same team of trained staff who followed identical standard operating procedures. Informed consent was obtained from all eligible participants and both studies gained full ethical and governance approvals from the Nottingham Research Ethics Committee (WA) and Leicestershire, Northamptonshire and Rutland Comprehensive Local Research Network (Project STAND).

Walking Away Middle aged and older adults (aged up to 74) were recruited from 10 primary care practices within Leicestershire, United Kingdom (UK). Individuals at high risk of impaired glucose regulation (IGR) (composite of impaired glucose tolerance (IGT) and/or impaired fasting glycaemia (IFG)) or type 2 diabetes mellitus were identified using a modified version of the automated Leicester Risk Score, specifically designed to be administered in primary care [21]. An automated platform using medical records was used to rank individuals for diabetes risk using predefined weighted variables (age, gender, BMI, family history of type 2 diabetes mellitus and use of antihypertensive medication). Those scoring within the 90^{th} percentile in each practice were invited to take part in the study. This approach has been shown to have reasonable sensitivity and specificity for identifying participants at a high risk of IGR [21].

Project STAND Young adults who were at risk of developing type 2 diabetes mellitus from across Leicestershire and the South East Midlands region were recruited from primary care practices. Practices databases were searched for participants meeting the following inclusion criteria: a) aged 18-40 years with a BMI in the obese range ($\geq$30kg/m^2; $\geq$27.5kg/m^2 for south Asians) or b) aged 18-40 years with a BMI in the overweight range $\geq$25kg/m^2 ($\geq$23kg/m^2 for south Asians) plus one additional risk factor; a family history of type 2 diabetes mellitus or cardiovascular disease (CVD), previous gestational diabetes, polycystic ovarian syndrome, HbA1c $\geq$5.8% or IGR [22].

Cardiovascular, metabolic and anthropometric outcomes Markers of metabolic and cardiovascular health were measured, including fasting plasma glucose (FPG) and 2-hour plasma glucose (via an OGTT), HbA1c, total cholesterol, HDL cholesterol and triacylglycerol. Venous blood samples were obtained following an overnight fast and all assays were measured in the same laboratory located within the Leicester Royal Infirmary, UK. Analysis was conducted by individuals blinded to the patients' identity, using stable methodologies, standardised to external quality assurance values. Plasma glucose was analysed in venous samples via the hexokinase method. HbA1c was analysed using the Bio-Rad Variant II HPLC system (Bio-Rad Clinical Diagnostics, Hemel Hempstead, UK). HDL-cholesterol and triacylglycerol were measured using standard enzymatic techniques.

Body weight (Tanita TBE 611, Tanita, West Drayton, UK) and waist circumference (midpoint between the lower costal margin and iliac crest) were measured to the nearest 0.1 kg and 0.5 cm respectively. Information on current smoking status, medication and ethnicity was obtained following an interview administered protocol with a health care professional. Social deprivation was determined by assigning an Index of Multiple Deprivation (IMD) score to the participant's resident area [23]. IMD scores are publically available continuous measures of compound social and material deprivation which are calculated using a variety of data including; current income, employment, education and housing.

Accelerometer measures Participants were asked to wear a tri-axial accelerometer (Actigraph GT3X, Florida, USA) on the right midaxillary line of the hip (attached via a waistband), for a minimum of seven consecutive days during waking hours. These accelerometers translate raw

accelerations into activity counts. Accelerometers were initialised to record activity in 5s epochs in the STAND cohort and 15s epochs in the WA cohort. During each sampling interval (5s or 15s), all registered activity counts were summed and stored in the monitor memory. In order to allow for direct comparison, all data from the STAND cohort were re-integrated into 15s epochs. Freedson cut-points were used to categorise each epoch as sedentary (<25 counts per 15 seconds), light-intensity physical activity (≥25 to <488 counts per 15 seconds), or MVPA (≥488 counts per 15 seconds) [24]. Breaks in sedentary time were defined as a transition from a sedentary (<25 counts per 15 seconds) to an active state (≥25 counts per 15 seconds) [4, 8]. Total physical activity counts represented the summation of counts within each epoch.

Non-wear time was defined as a minimum of 60 minutes of continuous zero counts and days with at least 600 minutes wear time were considered valid [4, 5, 14]. In order for data to be included in the analysis, participants required at least four valid days [25].

All accelerometer-derived variables (sedentary time, MVPA, breaks in sedentary time and total counts) were computed by summing the values over all valid days and calculating the mean value per valid day.

An accelerometer data analysis tool (ActiSCi, Suffolk, UK) was used to process the accelerometer data.

Statistical analysis Statistical analyses were performed using PASW Statistics v18.0. Due to their skewed distribution, FPG, 2-hour glucose, HDL-cholesterol, triacylglycerol and HDL: total cholesterol ratio were log-transformed.

Forced-entry linear regression analysis was used on the combined study cohorts to examine the independent associations of sedentary time, total physical activity, breaks in sedentary time and MVPA with markers of metabolic (FPG, 2-hour glucose, waist circumference, BMI, HbA1c) and cardiovascular health (triacylglycerol, HDL cholesterol and HDL: total cholesterol ratio).

Model 1 was adjusted for age, gender, smoking status, ethnicity, social deprivation, lipid lowering and beta-blocker medication, family history of type 2 diabetes mellitus and time accelerometer worn (average minutes per day). Model 2 was additionally adjusted for MVPA time (minutes per day) and the associations for breaks, MVPA and total physical activity were examined having also adjusted for sedentary time (minutes per day). In order to examine the extent to which adiposity may attenuate these relationships, model 3 was further adjusted for BMI.

Significant associations were followed up with interaction terms to assess differences in the strength of the associations between sedentary time, breaks in sedentary time, total physical activity and MVPA by study group and sex, using a model adjusted for the above covariates. To further represent the strength of sedentary time and breaks in sedentary time with cardio-metabolic markers, variables were also examined as tertiles using analysis of covariance (ANCOVA) procedures.

In order to enable direct comparison to previous published studies, a sensitivity analysis was conducted to investigate whether results were affected by integrating the measure of sedentary time to 60s epochs.

Two-tailed p values of 0.05 or less were considered statistically significant for main effects. Adjustment was not made for multiple comparisons, therefore data were viewed with caution and in relation to the overall pattern of results. $p<0.1$ was considered significant for interactions. Due to log-transformation, and to allow for direct comparisons across cardio-metabolic markers, results of the linear regression analysis are presented as the standardised beta co-efficient (β)±SE.

Results

In total, 153 younger participants from Project STAND (age=32.9±5.6years; female=71.2%) and 725 older participants from WA (age=63.7±7.8years; female=35.2%) had valid measures of both objective activity and biochemical variables. This equated to 87% of the combined cohort. The majority of excluded participants failed to meet the minimum accelerometer wear time requirement. Those included in this analysis had a similar ethnic breakdown and social deprivation score compared to those who did not reach the minimum accelerometer criteria. However, those excluded were more likely to be younger (51.3±14.6 vs. 58.4±13.8years, $p<0.001$), have a larger waist circumference (105.4±15.4 vs. 101.6±12.0cm, $p<0.001$) and higher BMI (34.6±6.7 vs. 32.5±5.2kg/m^2, $p<0.001$). Table 1 reports the demographic, cardio-metabolic, anthropometric and accelerometer characteristics of included participants.

Accelerometer wear time (Project STAND= 14.5±1.4 vs. WA=14.4±1.4hours per day) and sedentary time (10.3±1.5 vs. 10.3±1.5hours) were similar between study cohorts. The younger Project STAND cohort spent a longer time engaged in MVPA (interquartile range; 0.7 (0.4-0.9) vs. 0.5 (0.3-0.9hours)).

Sedentary time showed a moderate inverse correlation with total physical activity (r=-0.34, p=<0.001) and MVPA (r_s=-0.36, p=<0.001) and a small inverse correlation with breaks (r=-0.111, p=0.001). MVPA had a small association with breaks (r=0.23, p=<0.001) and was strongly correlated with total physical activity (r=0.88, p=<0.001). Furthermore, total physical activity displayed a moderate correlation with the number of breaks (r=0.31, p=<0.001)

Table 2 displays the adjusted associations in the combined cohort of sedentary time, total physical activity, MVPA and the number of breaks in sedentary time with biomedical and anthropometric markers.

Sedentary Time After adjustments for known confounders, including MVPA and BMI, sedentary time showed a detrimental association with 2-hour glucose (β=0.220±0.060, p=<0.001), HDL-cholesterol (β=-0.123±0.056, p=0.029) and triacylglycerol (β=0.206±0.061, p=0.001).

Total physical activity Total physical activity was inversely associated with a multitude of cardio-metabolic factors, including 2-hour glucose (β=-0.164±0.035, p=<0.001), waist circumference (β=-0.270±0.032, p=<0.001), BMI (β=-0.281±0.051, p=<0.001), triacylglycerol (β=-0.173±0.036, p=<0.001), total cholesterol:HDL ratio (β=-0.126±0.034, p=<0.001) and HDL cholesterol (β=0.160±0.033, p=<0.001). Associations with biochemical factors were weakened after further adjustment for sedentary time with only the association with HDL-cholesterol remaining significant. However, associations between total physical activity and measures of adiposity were largely unaffected by adjustment for sedentary time.

MVPA Time in MVPA was significantly inversely associated with 2-hour glucose (β=-0.121±0.035, p=<0.001), triacylglycerol (β=-0.149±0.036, p=<0.001), total cholesterol:HDL ratio (β=-0.124±0.034, p=<0.001), HDL cholesterol (β=0.150±0.033, p=<0.001), BMI (β=-0.241±0.031, p=<0.001) and waist circumference (β=-0.270±0.033, p=<0.001). However, after adjustment for sedentary time, only BMI (β=-0.215±0.041, p=<0.001) and waist circumference (β=-0.228±0.043, p=<0.001) remained significant.

Breaks in sedentary time Independent of known confounders (including sedentary time), breaks in sedentary time were significantly inversely associated with 2-hour glucose (β=-0.111±0.055, p=0.046) waist circumference (β=-0.215±0.051, p=<0.001) and BMI (β=-0.151±0.049, p=0.003). However, further adjustment for BMI attenuated the association with 2-hour glucose.

Results reported above were unaffected if waist circumference rather than BMI was used in Model 3.

Figure 1 illustrates the associations between sedentary time and 2-hour glucose, HDL-cholesterol and triacylglycerol when examined as tertiles. Figure 2 shows the association of breaks with waist circumference and BMI.

Interaction analyses indicated a significant effect for study cohort, with the older cohort demonstrating greater associations of MVPA and total physical activity with BMI (p for interaction <0.001) and waist circumference (p for interaction <0.001). For breaks in sedentary time, the same pattern was observed, with the older cohort achieving a stronger association for waist circumference (p for interaction=<0.001) and BMI (p for interaction=<0.001). No other significant interactions were observed for the effect of study group. In addition, there were no significant interactions for sex in the results for sedentary time, total physical activity, MVPA or breaks.

The pattern of results and significance levels were unaffected if data were analysed at 60s epochs. However, standardised beta-coefficients were consistently around 10% lower reflecting the less sensitive nature of the data at longer epochs (data available on request).

Discussion

This study demonstrates that for individuals with known risk factors for type 2 diabetes mellitus recruited from primary care, sedentary time was detrimentally associated with 2-hour glucose, triacylglycerol and HDL-cholesterol, independent of measured confounders. These results remained significant after further adjustment for measures of adiposity. Furthermore, the findings for biochemical factors were consistent across groups with diverse age ranges, providing evidence that the deleterious consequences of excess sedentary time exist across young to old adults. Interestingly, sedentary time was shown to have stronger associations with several important cardio-metabolic markers (2-hour glucose, triacylglycerol, HDL-cholesterol) compared to total physical activity and MVPA, after adjustment for each other and other important confounders. Associations of breaks in sedentary time with markers of health, independent of overall time spent sedentary and in MVPA, were less consistent, although beneficial associations were observed with measures of adiposity. To our knowledge, this is the first study to examine the effect of sedentary time and breaks on markers of cardio-metabolic health in a primary care population with known risk factors for type 2 diabetes mellitus.

Our study has multiple strengths; most notably it provides novel evidence in a high risk population recruited through primary care using an objective measure of sedentary time, across a wide age range. Furthermore, all participants were from the same geographical location, with similar risk profiles and measurements across both studies were performed by the same team of trained staff, following identical standard operating procedures. In addition, participants were rigorously phenotyped with traditional markers of cardio-metabolic health using standardised biochemical procedures. Limitations include the cross-sectional design, thus limiting inference about the direction of causality between the sedentary variables, physical activity and markers of cardio-metabolic health; reverse causality remains a possibility. Despite allowing for more robust assessments of sedentary behaviour compared to self-report, accelerometers are not without limitations. For example, they rely on categorising movement (acceleration) strength, rather than directly distinguishing between sitting, lying and standing behaviours. Furthermore, they may underestimate overall physical activity as they are unable to accurately quantify certain non-step based activities (i.e. cycling).

Our results extend those from other studies that have utilised both self-reported and objective measures of sedentary time and MVPA with cardio-metabolic variables in the general population. Self-reported sedentary behaviour in the form of television viewing time has been positively associated with a multitude of cardio-metabolic risk factors [6, 26-28], including 2-hour glucose [26, 27]. Similarly, recent reviews also report that self-reported sedentary time is consistently associated with an increased risk of diabetes [9] and metabolic syndrome [29].

Several studies have examined the joint effect of sedentary behaviour and physical activity on health outcomes [5, 30, 31]. In contrast to our observations, most have concluded that physical activity is a stronger predictor of metabolic risk [30] and insulin resistance [31]. This discrepancy in findings may be due to differences in study populations, as our participants had several known risk factors for type 2 diabetes mellitus and were largely obese. Indeed, our results are consistent with previous findings in a similar population, showing that sedentary time has stronger associations with various markers of cardio-metabolic health, compared to MVPA [32]. This is particularly important as our population is representative of those that are likely to be identified as being at high risk of type 2 diabetes mellitus within routine care and referred onto available prevention programmes. As such, these studies provide preliminary evidence that sedentary behaviour may be a more effective paradigm to target in the prevention of T2DM, rather than solely focusing on MVPA. Moreover, sedentary time takes up large portions of the day, unlike MVPA.

Despite a trend for higher levels of breaks to be associated with lower 2-hr glucose, our study was not able to corroborate a previous finding that breaks in sedentary time were independently associated with glucose regulation and triacylglycerol [8]. The discrepancy may be partly explained by the fact that our participants spent longer in sedentary pursuits and took fewer breaks compared to a similar population based study [8]. Nevertheless, our findings are broadly consistent with other studies conducted in the general population and in those with type 2 diabetes which showed no associations between breaks in sedentary time and measures of insulin resistance and lipid parameters [4, 32]. However, as with the present study, strong associations between breaks in sedentary time and measures of adiposity were observed. Consequently, this study further suggests that breaks in sedentary time, rather than total sedentary time *per se* may be an important factor in the regulation of body weight. This is consistent with a small intervention study which suggested that regular variations in posture allocation may be an influential factor in the regulation of energy homeostasis [33].

The non-significant associations observed for FPG and HbA1c across all measures of sedentary behaviour and physical activity are consistent with previous research [4, 14, 34] and reflect the different pathophysiological process underlying 2-hour and FPG regulation, with 2-hour glucose largely influenced by peripheral insulin resistance [34, 35]. Our findings, therefore, highlight the importance of using 2-hour glucose as the primary outcome variable when assessing the impact of sedentary time on cardio-metabolic risk.

Animal models have begun to elucidate the potential biological mechanisms that may underlie the relationship between sedentary behaviour and cardio-metabolic risk. Previous laboratory work has identified that distinctive physiological pathways are activated with increased sedentary behaviour, particularly around the metabolism of lipoprotein lipase, which remains largely unaffected by MVPA [36]. Lipoprotein lipase is a key regulator of lipid metabolism and is causally linked to CVD [37]. In addition, sedentary behaviour may also reduce glucose transporter protein content, thus exacerbating insulin resistance [38]. Nevertheless, published experimental research in humans is largely lacking and the underlying mechanisms are likely to be multifarious. In addition, there is a need to accumulate supplementary data from prospective studies and new evidence from human experimental work and intervention trials. To date, only one intervention study focused specifically on sedentary behaviour in adults has been published. Nineteen overweight/obese adults showed large reductions in the area under the glucose and insulin curve when sitting time was regularly punctuated with short periods of both light and moderate intensity activity [39]. Surprisingly, there was no difference between the effect sizes found in the light or moderate intensity profiles. Although encouraging, the findings from this study need to be confirmed in different populations in order to establish a causal link between sedentary behaviour and cardio-metabolic dysfunction.

In conclusion, the findings from this study may have important methodological and public health implications. This study provides novel objective evidence that in individuals at high risk of type 2 diabetes mellitus, sedentary time may be a more important indicator of cardio-metabolic health than MVPA. This may raise questions regarding the prescription of optimal daily human movement for health. As such, diabetes and cardiovascular prevention programmes concentrating solely on MVPA may overlook an area that is of fundamental importance to cardio-metabolic health. Along with messages around accumulating at least 150 minutes per week of MVPA, which form the corner-stone of diabetes prevention programmes [40], such interventions may be more effective if individuals are further encouraged to simply sit less and move more, regardless of the intensity level. This is an innovative approach which requires a paradigm shift, so that individuals think about the balance of sedentary behaviour and activity in all aspects of daily life. Nevertheless, given the limitations, this study should not be used to confirm a direct link between sitting time and metabolic health, but should act as a stimulus for tightly controlled intervention studies in different populations in order to influence future physical activity and sedentary behaviour interventions and public health initiatives aimed at disease prevention.

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Duality of interest

The authors declare that there is no duality of interest associated with this manuscript.

Author Contributions: JH and TY had original idea for the analysis. JH wrote the first draft of the article. JH, TY, SB, KK, EW, MN and MJD made substantial contributions to conception and design. JH and CE carried out acquisition of data. JH, CE and TG processed raw accelerometer files. JH, TY and LG carried out and analysis and interpretation of data. All authors reviewed/edited the manuscript and gave final approval of the version to be published.

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Tables

Table 1 Demographics, metabolic, anthropometric and accelerometer characteristics of participants

Characteristics	STAND (N=153)	Walking Away (N=725)	All (N=878)
Age (years)	32.9 ± 5.6	63.7 ± 7.8	58.4 ± 13.8
Male	44 (28.8)	470 (64.8)	514 (58.5)
Current smokers	57 (37.3)	62 (8.5)	119 (13.6)
Family history of diabetes (1st degree)	100 (65.4)	261 (35.9)	361 (41.1)
Cardio-metabolic variables			
BMI (kg/m^2)	34.6 ± 4.8	31.2 ± 5.3	32.5 ± 5.2
Waist circumference (cm)	102.9 ± 13.5	101.3 ± 11.7	101.6 ± 12.0
Weight (kg)	98.3 ± 17.3	91.5 ± 16.5	92.7 ± 16.9
FPG (mmol/l)	4.8 (4.5-5.1)	5.2 (4.9-5.6)	5.1 (4.8-5.5)
2-hour plasma glucose (mmol/l)	5.3 (4.5-6.4)	6.1 (4.9-7.8)	5.9 (4.8-7.5)
Body fat (%)	40.5 ± 7.2	35.6 ± 8.7	36.5 ± 8.6
Total cholesterol (mmol/l)[a]	4.8 (4.2-5.4)	5.1 (4.3-5.9)	5.0 (4.3-5.8)
HDL cholesterol (mmol/l)[a]	1.2 (1.0-1.4)	1.4 (1.1-1.6)	1.3 (1.1-1.6)
Total cholesterol: HDL ratio (mmol/l)[a]	3.9 (3.2-4.7)	3.7 (3.0-4.4)	3.8 (3.0-4.5)
Triacylglycerol (mmol/l)[a]	1.30 (0.90-1.70)	1.30 (1.00-1.80)	1.30 (1.00-1.80)
Lipid lowering medication	1 (0.6)	240 (33.1)	241 (27.4)
Beta-blockers	2 (1.3)	127 (15.2)	129 (14.7)
HbA1c (%)	5.6 (5.3-5.8)	5.9 (5.6-6.1)	5.8 (5.6-6.1)
Ethnicity			
White European	128 (83.7)	645 (89.0)	773 (88.0)
South Asian	15 (9.8)	53 (7.3)	68 (7.8)
Other	10 (6.5)	27 (3.7)	37 (4.2)
Diagnosis			
Normal glucose tolerance	137 (89.5)	512 (70.6)	649 (73.9)
Isolated IFG	3 (1.9)	38 (5.2)	41 (4.7)
Isolated IGT	11 (7.2)	124 (17.1)	135 (15.4)
Both	1 (0.7)	31 (4.3)	32 (3.6)
Type 2 diabetes mellitus	1 (0.7)	20 (2.8)	21 (2.4)
All (IGR)	16 (10.4)	214 (29.5)	230 (26.2)
Accelerometer variables (time in hours)			
Time accelerometer worn	14.5 ± 1.4	14.4 ± 1.4	14.4 ± 1.4
Sedentary time[b]	10.3 ± 1.5	10.3 ± 1.5	10.3 ± 1.5
Light-intensity activity[b]	3.5 ± 0.9	3.5 ± 0.9	3.5 ± 0.9
MVPA[b]	0.7 (0.4-0.9)	0.5 (0.3-0.9)	0.6 (0.3-0.9)
Accelerometer variables (percent at each activity level)			
Sedentary time	70.5 ± 7.6	71.5 ± 7.8	71.0 ± 8.0
Light-intensity activity	24.3 ± 6.5	24.3 ± 6.3	24.3 ± 6.4
MVPA	4.7 (2.8-6.2)	3.7 (2.1-5.9)	3.9 (2.3-6.0)
Breaks per day	297 ± 68	273 ± 60	277 ± 62
Total physical activity counts (x 1000·day)	274 ± 109	253 ± 126	257 ± 123
Average steps per day	7153 ± 2954	6993 ± 3384	7016 ± 3313

[a]Means adjusted for lipid lowering medication
[b]Means adjusted for the time the accelerometer was worn
Sedentary time = <25 counts per 15s, light intensity activity ≥25 to <488 counts per 15s, and MVPA ≥488 counts per 15s. Continuous parametric results as mean±SD, number (column percentage) and continuous non-parametric results as median (interquartile range)

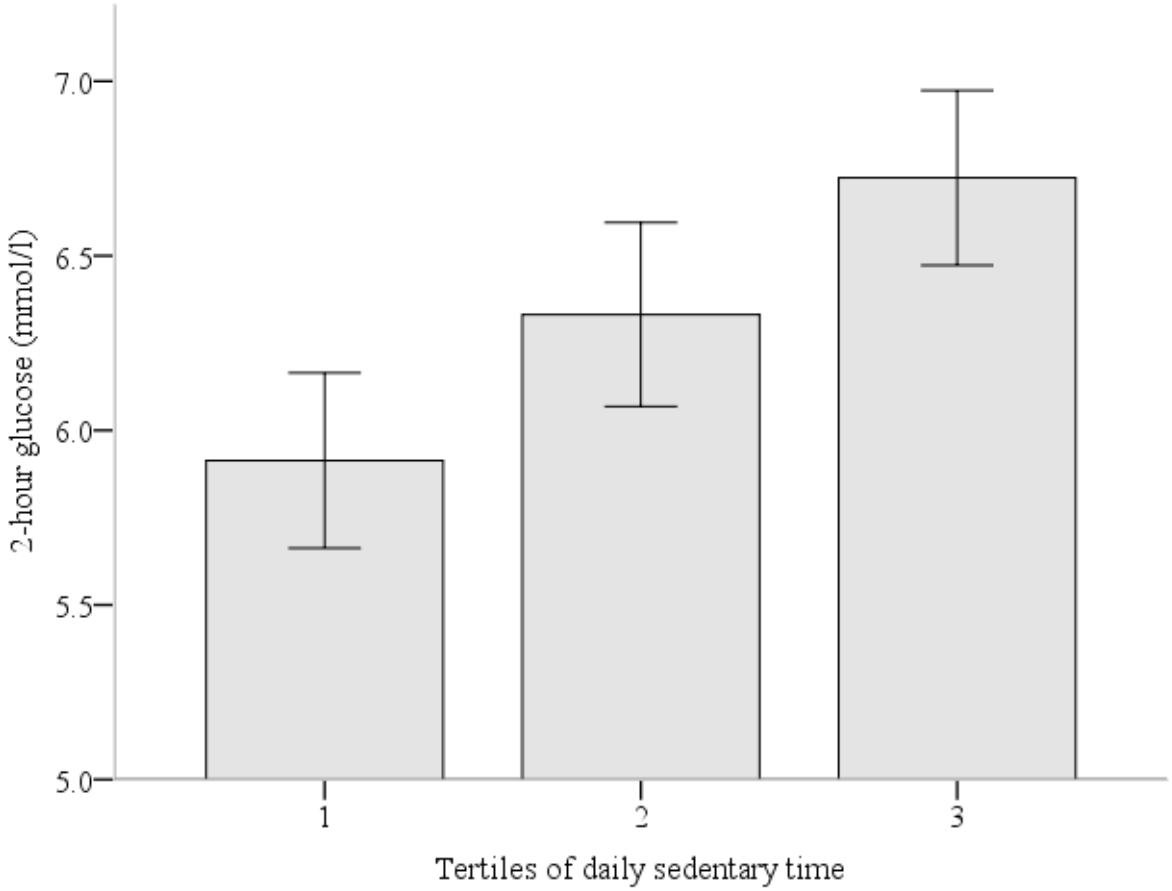
Table 2 Multiple linear regression models for sedentary time, total physical activity, moderate-to-vigorous physical activity and breaks in sedentary time with cardio-metabolic variables

Model 1								
	Sedentary time (<25 counts per 15s) *β* (SE)	*p* value	Total physical activity (cpm) *β* (SE) [b]	*p* value	MVPA (≥488 counts per 15s) *β* (SE)	*p* value	Breaks *β* (SE)	*p* value
2-hour glucose (mmol/l)	0.238 (0.045)	<0.001	-0.164 (0.035)	<0.001	-0.121 (0.035)	0.001	-0.180 (0.038)	<0.001
Waist circumference (cm)	0.250 (0.043)	<0.001	-0.270 (0.032)	<0.001	-0.270 (0.033)	<0.001	-0.198 (0.037)	<0.001
BMI (kg/m^2)	0.210 (0.041)	<0.001	-0.281 (0.051)	<0.001	-0.241 (0.031)	<0.001	-0.148 (0.035)	<0.001
Triacylglycerol (mmol/l)	0.217 (0.045)	<0.001	-0.173 (0.036)	<0.001	-0.149 (0.036)	<0.001	-0.150 (0.040)	<0.001
Fasting glucose (mmol/l)	0.046 (0.045)	0.308	-0.068 (0.058)	0.248	-0.033 (0.036)	0.488	-0.024 (0.038)	0.777
Total cholesterol:HDL ratio	0.130 (0.043)	0.003	-0.126 (0.034)	<0.001	-0.124 (0.034)	<0.001	-0.114 (0.037)	0.003
HDL cholesterol (mmol/l)	-0.187 (0.042)	<0.001	0.160 (0.033)	<0.001	0.150 (0.033)	<0.001	0.130 (0.036)	<0.001
HbA1c (%)	0.035 (0.050)	0.489	0.031 (0.035)	0.379	-0.034 (0.046)	0.464	-0.021 (0.038)	0.590
Model 2								
	Sedentary time (<25 counts per 15s) *β* (SE) [a]	*p* value	Total physical activity (cpm) *β* (SE) [b]	*p* value	MVPA (≥488 counts per 15s) *β* (SE) [b]	*p* value	Breaks *β* (SE) [c]	*p* value
2-hour glucose (mmol/l)	0.235 (0.060)	<0.001	-0.038 (0.073)	0.494	-0.033 (0.047)	0.473	-0.111 (0.055)	0.046
Waist circumference (cm)	0.091 (0.057)	0.113	-0.259 (0.070)	<0.001	-0.228 (0.043)	<0.001	-0.215 (0.051)	<0.001
BMI (kg/m^2)	0.054 (0.053)	0.327	-0.247 (0.055)	<0.001	-0.215 (0.041)	<0.001	-0.151 (0.049)	0.003
Triacylglycerol (mmol/l)	0.214 (0.062)	0.001	-0.067 (0.060)	0.266	-0.042 (0.048)	0.385	-0.046 (0.056)	0.418
Fasting glucose (mmol/l)	0.023 (0.062)	0.714	-0.040 (0.035)	0.257	-0.021 (0.048)	0.662	-0.011 (0.038)	0.903
Total cholesterol:HDL ratio	0.101 (0.058)	0.085	-0.085 (0.070)	0.120	-0.075 (0.045)	0.096	-0.075 (0.054)	0.167
HDL cholesterol (mmol/l)	-0.137 (0.056)	0.016	0.120 (0.052)	0.022	0.083 (0.044)	0.060	0.071 (0.052)	0.175
HbA1c (%)	0.014 (0.051)	0.836	0.024 (0.056)	0.670	-0.013 (0.036)	0.725	-0.035 (0.056)	0.537
Model 3								
	Sedentary time (<25 counts per 15s) *β* (SE) [a]	*p* value	Total physical activity (cpm) *β* (SE) [b]	*p* value	MVPA (≥488 counts per 15s) *β* (SE) [b]	*p* value	Breaks *β* (SE) [c]	*p* value
2-hour glucose (mmol/l)	0.220 (0.060)	<0.001	-0.017 (0.057)	0.766	-0.019 (0.055)	0.678	-0.095 (0.056)	0.091
Triacylglycerol (mmol/l)	0.206 (0.061)	0.001	-0.021 (0.061)	0.732	-0.011 (0.050)	0.826	-0.019 (0.056)	0.736
Fasting glucose (mmol/l)	0.011 (0.063)	0.857	-0.023 (0.033)	0.694	-0.009 (0.047)	0.850	0.000 (0.050)	0.993
Total cholesterol:HDL ratio	0.090 (0.057)	0.120	-0.033 (0.055)	0.547	-0.037 (0.045)	0.412	-0.044 (0.053)	0.408
HDL cholesterol (mmol/l)	-0.123 (0.056)	0.029	0.063 (0.052)	0.228	0.041 (0.043)	0.344	0.035 (0.051)	0.495
HbA1c (%)	0.008 (0.062)	0.898	0.064 (0.067)	0.260	-0.010 (0.046)	0.828	-0.022 (0.054)	0.689

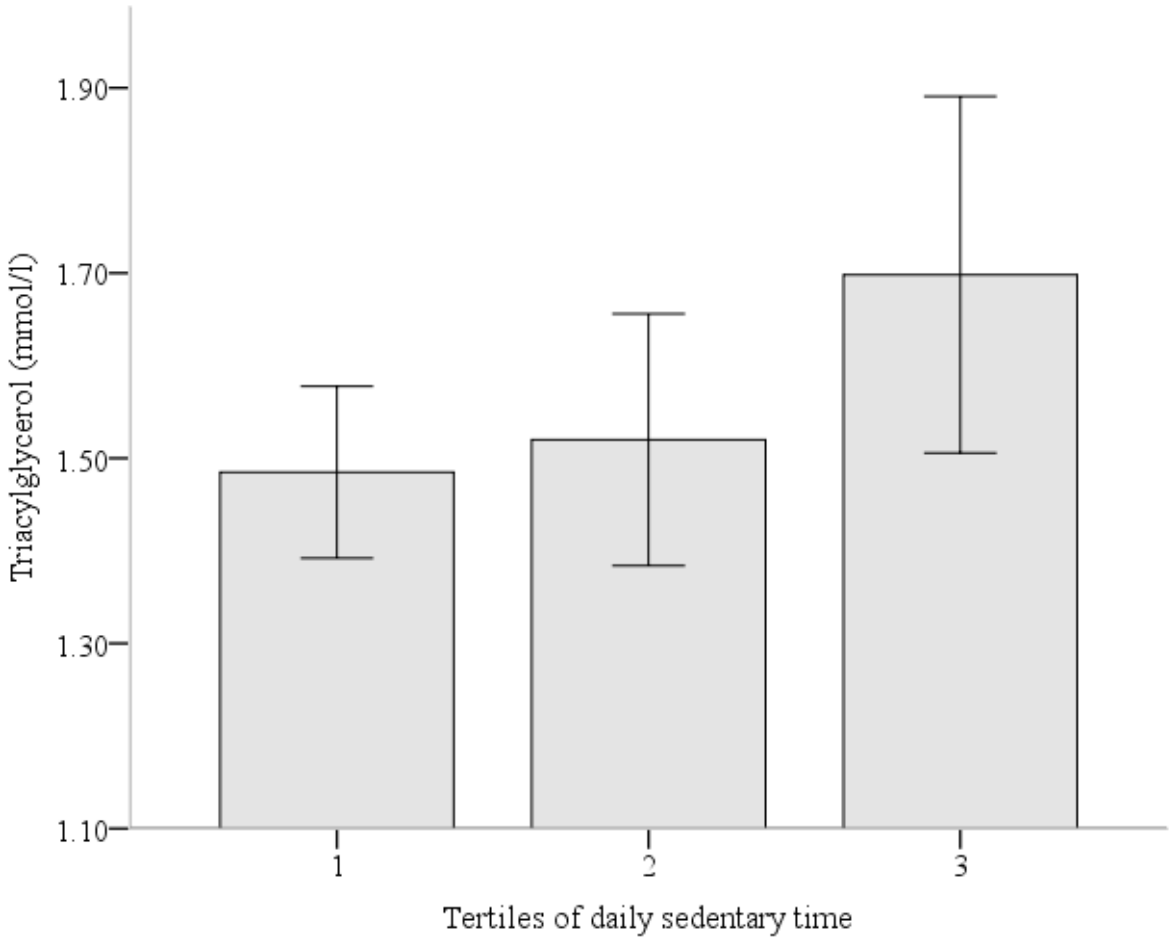
Model 1 was adjusted for age, gender, smoking status, ethnicity, social deprivation, family history, beta blockers, lipid lowering medication and time accelerometer worn
Model 2 was adjusted for the above covariates and [a]MVPA, [b]sedentary time or [c]sedentary time and MVPA
Model 3 was adjusted for the same covariates as Model 2 and BMI

Figures

a


7.0
6.5
6.0
5.5
5.0
2-hour glucose (mmol/l)
1
2
3
Tertiles of daily sedentary time

b


1.90
1.70
1.50
1.30
1.10
Triacylglycerol (mmol/l)
1
2
3
Tertiles of daily sedentary time

c

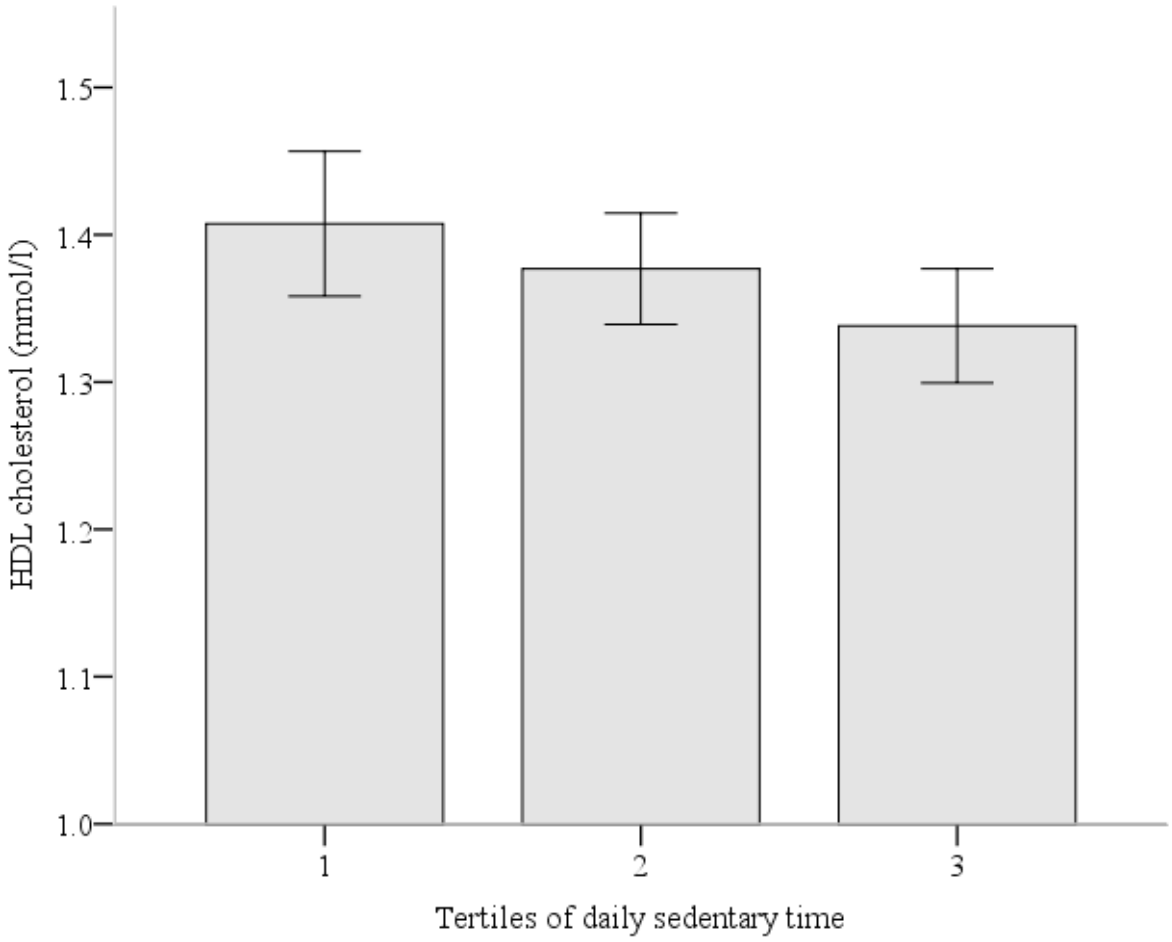
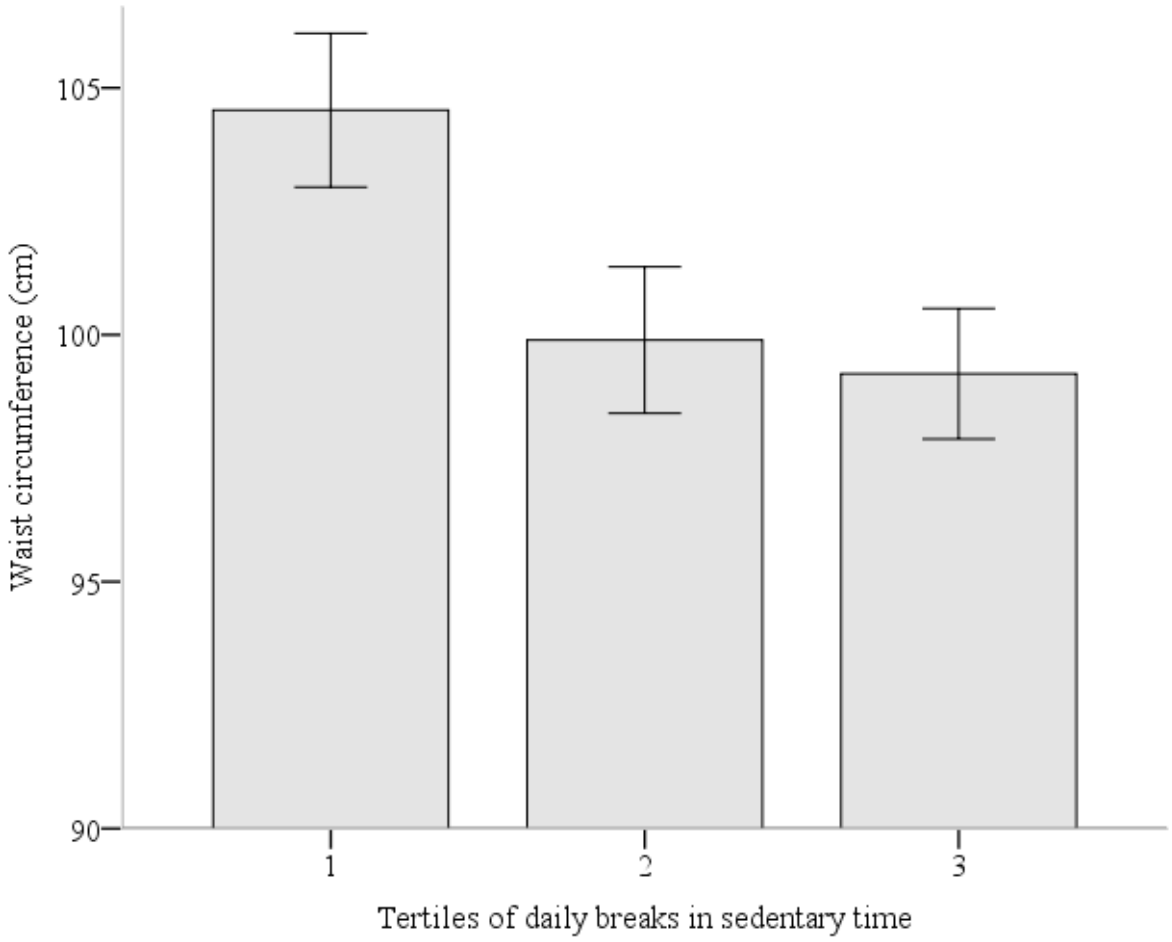


Figure 1 Tertiles of sedentary time with 2-hour glucose (a), triacylglycerol (b) and HDL-cholesterol (c). Estimated marginal means are adjusted for age, gender, ethnicity, IMD score, smoking status, family history of type 2 diabetes mellitus, lipid lowering medication, beta blockers, time accelerometer worn, days accelerometer worn, time spent in MVPA and BMI. Tertile cut-points for sedentary time were 9.6 and 10.9hours/day. Medians and ranges for tertile 1 = 8.7hours (2.9-9.5); tertile 2= 10.3hours (9.6-10.9); tertile 3= 11.7hours (11.0-15.8). $p<0.001$ for trend (a), $p<0.05$ for trend (b, c).

a


105
100
95
90
Waist circumference (cm)
1
2
3
Tertiles of daily breaks in sedentary time

b

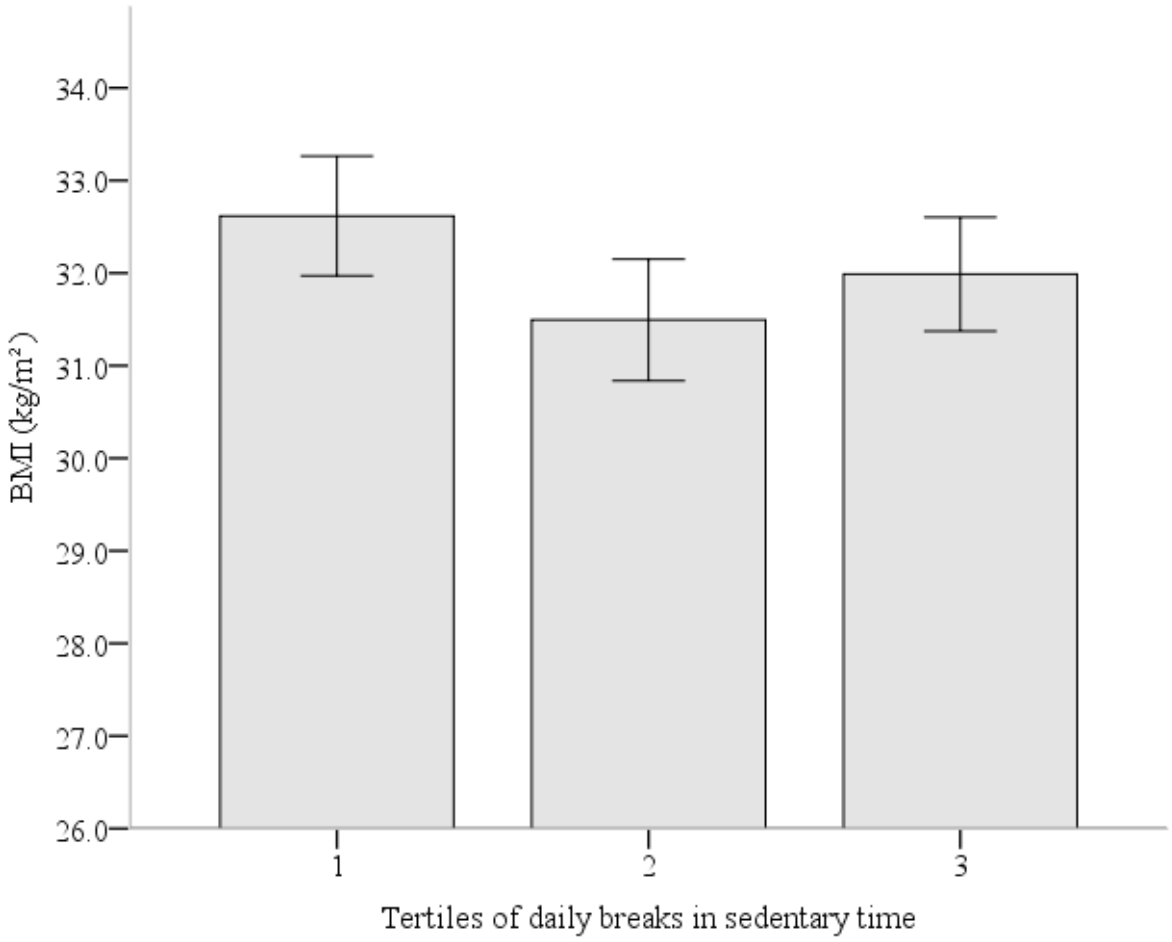


Figure 2 Tertiles of breaks in sedentary time with waist circumference (a) and BMI (b). Estimated marginal means are adjusted for age, gender, ethnicity, IMD score, smoking status, family history of type 2 diabetes mellitus, lipid lowering medication, beta blockers, time accelerometer worn, days accelerometer worn and time spent in sedentary and MVPA. Cut points for daily breaks in sedentary time were 234 and 285. Medians and ranges for tertile 1 = 215 (33-234); tertile 2= 268 (235-284); tertile 3= 329 (285-487). $p<0.001$ for trend (a), $p<0.01$ for trend (b)