

Replacing sitting time with standing or stepping: associations with cardio-metabolic risk biomarkers

Genevieve N. Healy^{1,2,3*}, Elisabeth A. H. Winkler¹, Neville Owen^{1,2,4,5,6}, Satyamurthy Anuradha¹, and David W. Dunstan^{1,2,4,7,8,9,10,11}

¹School of Public Health, The University of Queensland, Herston Road, Herston, Brisbane, QLD 4006, Australia; ²Baker IDI Heart and Diabetes Institute, Melbourne, VIC, Australia; ³School of Physiotherapy, Curtin University, Perth, WA, Australia; ⁴Department of Medicine, Monash University, Malvern East, VIC, Australia; ⁵Department of Epidemiology and Preventive Medicine, Monash University, Malvern East, VIC, Australia; ⁶School of Population Health, The University of Melbourne, Melbourne, VIC, Australia; ⁷Department of Physiology, Monash University, Melbourne, VIC, Australia; ⁸Department of Physiology, The University of Melbourne, Melbourne, VIC, Australia; ⁹Centre for Physical Activity and Nutrition Research, School of Exercise and Nutrition Sciences, Burwood, VIC, Australia; ¹⁰School of Sport Science, Exercise and Health, The University of Western Australia, Perth, WA, Australia; and ¹¹Mary MacKillop Institute for Health Research, The Australian Catholic University, Melbourne, VIC, Australia

Received 31 March 2015; revised 30 May 2015; accepted 17 June 2015; online publish-ahead-of-print 30 July 2015

See page 2650 for the editorial comment on this article (doi:10.1093/eurheartj/ehv356)

Aims

While excessive sitting time is related adversely to cardio-metabolic health, it is unknown whether standing is a suitable replacement activity or whether ambulatory movement is required. Using isotemporal substitution analyses, we modelled cross-sectional associations with cardio-metabolic risk biomarkers of reallocating time (2 h/day) from sitting to standing or to stepping.

Methods and results

A subsample of participants from the 2011/12 Australian Diabetes, Obesity, and Lifestyle Study wore the posture-based activPAL3 monitor [36–80 years (mean 57.9, SD 9.9 years); 57% women; $n = 698$ with data]. Associations of activPAL3-derived mean daily time sitting/lying (sitting), standing and stepping with body mass index (BMI), waist circumference, blood pressure, HbA_{1c}, fasting glucose and lipids (high-density lipoprotein-, HDL, and low-density lipoprotein-cholesterol, total/HDL-cholesterol ratio, and triglycerides), and 2-h plasma glucose were examined. Adjusted for relevant confounders, sitting-to-standing reallocations were only significantly ($P < 0.05$) associated with approximately 2% lower fasting plasma glucose, 11% lower triglycerides, 6% lower total/HDL-cholesterol ratio, and 0.06 mmol/L higher HDL-cholesterol per 2 h/day. Sitting-to-stepping reallocations were only significantly associated with approximately 11% lower BMI, 7.5 cm lower waist circumference, 11% lower 2-h plasma glucose, 14% lower triglycerides, and 0.10 mmol/L higher HDL-cholesterol per 2 h/day, while standing-to-stepping reallocations were only significantly associated with ~10% lower BMI, 7 cm lower waist circumference, and 11% lower 2-h plasma glucose.

Conclusion

Findings suggested that sitting-reduction strategies targeting increased standing, stepping, or both, may benefit cardio-metabolic health. Standing is a simple alternative to sitting, and requires further examination in prospective and intervention studies.

Keywords

Sitting • Standing • Isotemporal • Cross-sectional • Adult • Cardio-metabolic

Clinical perspective

- Standing can be a feasible alternative to sitting in many contexts.
- Cardiovascular health benefits—particularly for glucose and lipid metabolism—may be achieved by reducing sitting through standing.
- For addressing overweight and obesity, sitting may need to be replaced with ambulatory movement.

* Corresponding author. Tel: +61 7 3365 5039, Fax: +61 7 3365 5540, Email: g.healy@uq.edu.au

Published on behalf of the European Society of Cardiology. All rights reserved. © The Author 2015. For permissions please email: journals.permissions@oup.com.

Introduction

High levels of sedentary time—or, *too much sitting*—have been linked detrimentally with cardiovascular disease, diabetes, and premature mortality.^{1,2} In modern society, adults are highly sedentary, with the average self-reported sitting time ranging from 3.2 to 6.8 h/day across 32 European countries³ and objective measures indicating 55–69% of adults' waking hours are spent sedentary.^{4,5} Accordingly, broadly stated guidelines on reducing sitting time have emerged.^{6,7} For these to become more specific guides to action, it is important to understand the relative benefits of the common daily activities (standing and stepping) that could replace sitting. The cardio-metabolic health benefits of ambulatory activity are well established.⁸ However, the potential benefits (or harms) of standing, a non-ambulatory alternative to sitting, are less well understood. Experimental studies have shown acute benefits of standing for postprandial glucose responses,^{9,10} but there is little evidence regarding non-acute relationships of directly measured standing with glucose metabolism or other cardio-metabolic risk biomarkers. Moreover, existing evidence seldom considers time displacement—that reduced time spent in one activity (e.g. sitting) inevitably increases time spent in other activities (e.g. standing). The potential cardio-metabolic impact of sitting reductions should include both the impact of reducing sitting time and of increasing time spent in non-sitting activities (i.e. standing or stepping).

Isotemporal substitution addresses such time displacements, estimating associations observed when cross-sectionally reallocating time from one activity to another, keeping total time and time in other activities fixed.¹¹ Studies measuring activity with hip-worn accelerometers suggest the benefits of reducing sedentary time on cardio-metabolic biomarkers likely vary depending on what displaces sedentary time. Stronger associations are observed when time (e.g. 1 h) is reallocated from sedentary to moderate- to vigorous-intensity activity than to light-intensity activity.^{12,13} However, isotemporal substitution has not yet been used to examine the potential health-related impacts of reallocating time from sitting to standing. This is necessary given recent findings show large shifts between these non-ambulatory activities are feasible and acceptable¹⁴ but have an unknown cardio-metabolic impact.

Using data collected from postural sensors, we examined cross-sectional associations of sitting, standing, and stepping with cardio-metabolic risk biomarkers in a broad sample of Australian adults. Associations were estimated considering time displacement (isotemporal substitution).

Research Design and Methods

Study design, participants, and recruitment

The Australian Diabetes, Obesity, and Lifestyle Study (AusDiab), a general population-based sample of community-dwelling Australian adults aged ≥ 25 initiated in 2000, had its third wave of data collection in 2011/12 (AusDiab3). This included objective activity assessment in a subsample of eligible participants recruited from on-site attendees ($n = 4614$) from 46 sites across Australia.¹⁵ Participants were invited consecutively until either no more devices were

available or five participants had been recruited for that day. A total of 1014 participants were approached; of these, 782 agreed to wear the activity monitor and 741 (73%) of these provided at least 1 day of valid data. The study complies with the Declaration of Helsinki. Ethics was approved by the Alfred Health Human Ethics Committee. Written informed consent was obtained.

Data collection

On the day of recruitment, as part of the AusDiab study procedures (protocols previously published^{15,16}), participants underwent biochemical, anthropometric, and behavioural assessments. In brief, following an overnight fast, a standard oral glucose tolerance test was performed, during which time all other data were collected, and the activity monitors were attached.

Measures

Activity outcomes

Activity outcomes were measured by the highly accurate^{17–20} activPAL3 activity monitor (PAL Technologies Limited, Glasgow, UK; version 6.4.1; see Supplementary material online, *Table S1*). The monitor was initialized, waterproofed, and then secured onto the right anterior thigh with a hypoallergenic patch. Participants were asked to wear the monitor continuously (24 h/day) for 7 days following the onsite assessment and to report in a diary all wake up, sleep ('lights out'), and monitor removal times (if any). Monitor data were processed in SAS 9.3 (SAS Institute Inc., Cary, NC, USA; see Supplementary material online, *Table S1*). Periods spent sleeping or not wearing the monitor and invalid days were excluded. For each participant, MET (metabolic equivalent)-minutes of stepping and time spent sitting, standing, stepping, and stepping at moderate- to vigorous-intensity physical activity (MVPA; ≥ 3 METs) were totalled for each day, averaged across valid days, then intensity (METs) of stepping time was calculated (MET-minutes/minutes of stepping).

Cardio-metabolic and anthropometric outcomes

Blood was collected via venepuncture and analysed at a central laboratory in Melbourne, Victoria (Healthscope Pathology). Serum triglycerides, total cholesterol, and high-density lipoprotein (HDL) cholesterol were measured by enzymatic methods; fasting and 2-h post-load plasma glucose were measured via a hexokinase method (Siemens Advia 2400). Low-density lipoprotein (LDL) cholesterol was determined using the Friedewald equation.²¹ Height (stadiometer) and weight (digital scales) were measured without shoes to the nearest 0.5 and 0.1 kg, respectively. Blood pressure was measured in triplicate in the seated position after rest for ≥ 5 min using an automated blood pressure monitor (Dinamap[®] Pro-series Monitor Model DP 101-NIBP). Minimum differences of interest (per 2 h/day of activity) were: 5% body mass index (BMI); 2 cm waist circumference; 5 mmHg systolic blood pressure; 3 mmHg diastolic blood pressure; 10% fasting glucose, post-load glucose and HbA1c; 5% total-, HDL-, LDL-cholesterol, and total/HDL-cholesterol ratio (equivalent to 0.26 mmol/L total cholesterol, 0.08 mmol/L HDL-cholesterol, and 0.15 mmol/L LDL-cholesterol); and 10% triglycerides.

Demographic and behavioural attributes

Socio-demographic and behavioural attributes, medical history, and current use of antihypertensive, diabetes, and lipid lowering medication were assessed via interviewer-administered questionnaires and categorized as per Supplementary material online, *Table S2*. Dietary variables were assessed via the self-completed Dietary Questionnaire for Epidemiological Studies Version 2.²²

Analysis

Statistical analyses were performed in STATA version 12 (StataCorp LP, College Station, TX, USA), using linearized variance estimation (survey commands) to account for the cluster design of the AusDiab study. Significance was set at a two-tailed $P < 0.05$ and to $P < 0.001$ for interactions (due to the excessive number of these assumption tests). Participants who provided at least one valid day of monitor data ($n = 741$) were not pregnant ($n = 739$), and who had complete data on covariates and outcomes ($n = 698$ or $n = 664$ for post-load glucose, which was not assessed in those taking diabetes medications) were included.

The characteristics of third-wave AusDiab attendees who were/were not included (due to ineligibility, sampling or non-participation; Supplementary material online, *Table S2*) and the baseline (1999/2000) characteristics of those who attended/did not attend the third AusDiab wave (Supplementary material online, *Table S3*) were compared using logistic regression. Associations of activities with cardio-metabolic biomarkers were examined using linear regression models. Sitting, standing, and stepping were considered individually, adjusting for waking wear time (residuals method) and potential confounders (Model A), and also further adjusting for MVPA (stepping at ≥ 3 METs; Model B). Potential confounders were age, gender, and any characteristic that showed evidence of association with the outcome ($P < 0.2$ in backward elimination; Supplementary material online, *Table S4*). When significant associations were observed, the interdependent (time displacement) associations were then tested using isotemporal substitution.¹¹ Associations were reported as regression coefficients or relative rates for log-transformed outcomes, with 95% confidence intervals, per 2 h/day reallocated. These indicate the cross-sectional associations with mean levels of biomarkers observed with reducing the mean time spent in less active behaviours (i.e. sitting or standing) by equivalently increasing the mean time spent in more active behaviours (i.e. standing or stepping) by 2 h/day without altering total waking wear time.

Models reported did not display problems with non-normality, heteroscedasticity, or non-linearity; all variance inflation factors were < 2.5 (Supplementary material online, *Table S5*). Partition models displayed multicollinearity; therefore, these were not reported. Sleep time (which has non-linearity issues) was not included in the models; only reallocations between waking activities were considered.

Results

Sample characteristics

Socio-demographic, behavioural, and health characteristics of participants are provided in Supplementary material online, *Table S2*. The sample covered ages 36–80 (median = 57) years, with 57% women.

Most participants provided at ≥ 4 days of monitor data ($n = 678$, 97%) and many provided all 7 ($n = 572$, 82%). On average (mean \pm SD), worn waking hours (15.7 ± 1.1 h/day) were mostly spent sitting (8.8 ± 1.8 h/day) and standing (4.9 ± 1.5 h/day), with 2.0 ± 0.7 h of stepping and 1.2 ± 0.4 h of MVPA per day. Participants' stepping intensity was on average 3.11 ± 0.16 METs.

Selection bias

Minor, but statistically significant, differences between the included substudy participants ($n = 698$) and the other AusDiab wave three attendees ($n = 3916$) were observed (Supplementary material online, *Table S2*). There was a tendency to exclude those with lower dietary fat intakes and those who were older, shorter, of lower socioeconomic position, post-menopausal, not taking the oral contraceptive pill, and with some poorer health characteristics. As previously observed,¹⁵ there were some biases in loss to follow-up with a number of small but statistically significant differences in baseline characteristics between those who attended the wave three follow-up vs. those who did not (Supplementary material online, *Table S3*).

Associations of sitting, standing, and stepping with cardio-metabolic biomarkers

The associations with cardio-metabolic biomarkers of sitting, standing, and stepping examined individually are shown in *Table 1*. Adjusted for confounders, each additional 2 h/day spent sitting (i.e. 2 h/day less standing and stepping) was significantly associated with higher BMI ($\approx 3\%$ with RR = 1.03, 95% CI: 1.01, 1.05), waist circumference ($\beta = 2.12$, 95% CI: 0.83, 3.41, i.e. ≈ 2 cm), fasting plasma glucose ($\approx 1\%$), total/HDL-cholesterol ratio ($\approx 5\%$), triglycerides ($\approx 12\%$), 2-h plasma glucose ($\approx 4\%$), and lower HDL-cholesterol (≈ 0.07 mmol/L). The associations of sitting with fasting glucose and the lipids were independent of MVPA, while associations with the adiposity markers and 2-h plasma glucose were attenuated in magnitude and no longer statistically significant following MVPA adjustment. Conversely, each 2 h/day spent standing was significantly associated with lower fasting plasma glucose ($\approx 2\%$ based on 1/RR with RR = 0.98), total/HDL-cholesterol ratio ($\approx 6\%$), triglycerides ($\approx 14\%$), and 2-h plasma glucose ($\approx 3\%$) and higher HDL-cholesterol (≈ 0.07 mmol/L). Only the association of standing with 2-h plasma glucose was attenuated with adjustment for MVPA (to $\approx 2\%$, $P = 0.104$). Each 2 h per day of stepping was significantly associated with lower BMI ($\approx 11\%$), waist circumference (≈ 8 cm), total/HDL-cholesterol ratio ($\approx 6\%$), triglycerides ($\approx 20\%$), and 2-h plasma glucose ($\approx 14\%$) and higher HDL-cholesterol (≈ 0.14 mmol/L). None of the activities showed significant associations with systolic and diastolic blood pressure, HbA_{1c} or LDL-cholesterol. The only inconclusive non-significant findings were for associations of standing with waist circumference, and for stepping with diastolic blood pressure and LDL-cholesterol (with confidence intervals containing meaningful associations, based on the minimum differences of interest). Associations of sitting, standing, and stepping with cardio-metabolic biomarkers did not vary significantly by age or gender at $P < 0.001$ (Supplementary material online, *Table S6*).

Table 1 Associations of objective, posture-based measures of sitting, standing, and stepping time with cardio-metabolic biomarkers in Australian adults >35 years (*n* = 698)

	Model ^a	Sitting (2 h/day)		Standing (2 h/day)		Stepping (2 h/day)	
		β or RR (95%CI)	<i>P</i>	β or RR (95%CI)	<i>P</i>	β or RR (95%CI)	<i>P</i>
BMI (kg/m ²), RR ^b	A	1.03 (1.01, 1.05)	0.002	0.98 (0.96, 1.00)	0.065	0.90 (0.86, 0.94)	<0.001
	B	1.01 (1.00, 1.03)	0.142	0.99 (0.97, 1.01)	0.184	–	–
Waist circumference (cm), β	A	2.12 (0.83, 3.41)	0.002	–1.48 (–3.09, 0.13)	0.071	–7.88 (–10.98, –4.79)	<0.001
	B	0.92 (–0.39, 2.24)	0.164	–0.99 (–2.47, 0.49)	0.183	–	–
Systolic BP (mmHg), β	A	–0.76 (–1.87, 0.35)	0.173	0.96 (–0.32, 2.25)	0.136	0.75 (–3.01, 4.50)	0.689
	B	–0.83 (–2.08, 0.42)	0.185	0.94 (–0.38, 2.27)	0.158	–	–
Diastolic BP (mmHg), β	A	0.56 (–0.21, 1.32)	0.151	–0.49 (–1.45, 0.48)	0.314	–1.51 (–3.83, 0.80)	0.193
	B	0.42 (–0.49, 1.32)	0.355	–0.42 (–1.40, 0.56)	0.390	–	–
Fasting plasma glucose (mmol/L), RR ^b	A	1.01 (1.00, 1.03)	0.039	0.98 (0.96, 1.00)	0.038	0.97 (0.94, 1.01)	0.142
	B	1.02 (1.00, 1.03)	0.033	0.98 (0.97, 1.00)	0.040	–	–
HbA _{1c} (mmol/mol), RR ^b	A	1.00 (1.00, 1.01)	0.350	1.00 (0.99, 1.01)	0.383	0.99 (0.97, 1.02)	0.555
	B	1.00 (1.00, 1.01)	0.371	1.00 (0.99, 1.01)	0.405	–	–
Total/HDL-cholesterol ratio, RR ^b	A	1.05 (1.03, 1.07)	<0.001	0.94 (0.92, 0.96)	<0.001	0.94 (0.90, 0.98)	0.008
	B	1.06 (1.03, 1.08)	<0.001	0.94 (0.92, 0.96)	<0.001	–	–
HDL-cholesterol (mmol/L), β	A	–0.07 (–0.10, –0.04)	<0.001	0.07 (0.04, 0.11)	<0.001	0.14 (0.07, 0.22)	0.001
	B	–0.06 (–0.09, –0.03)	0.001	0.07 (0.03, 0.10)	<0.001	–	–
LDL-cholesterol (mmol/L), β	A	0.03 (–0.03, 0.08)	0.294	–0.06 (–0.13, 0.01)	0.078	0.11 (–0.03, 0.25)	0.119
	B	0.06 (–0.01, 0.12)	0.087	–0.07 (–0.14, 0.00)	0.059	–	–
Triglycerides (mmol/L), RR ^b	A	1.12 (1.08, 1.15)	<0.001	0.88 (0.85, 0.92)	<0.001	0.83 (0.75, 0.91)	<0.001
	B	1.11 (1.07, 1.15)	<0.001	0.89 (0.86, 0.93)	<0.001	–	–
2-h post-load glucose (mmol/L), RR ^{b,c}	A	1.04 (1.01, 1.06)	0.003	0.97 (0.95, 1.00)	0.039	0.88 (0.83, 0.94)	<0.001
	B	1.02 (1.00, 1.04)	0.096	0.98 (0.95, 1.00)	0.104	–	–

^aModel A adjusted for age, gender, and confounders (backward elimination; Supplementary material online, Table S4). Model B adjusted for all variables in Model A and MVPA (≥ 3 METs) stepping. Time spent sitting, standing, and stepping are adjusted for wear time using the residuals method.

^bLog-transformed outcome; regression coefficients (β) and confidence intervals are back-transformed, exp (β), as relative rate (RR) with 95% confidence interval (CI).

^c'No difference' is indicated by $\beta = 0$ and RR = 1.

^dModels exclude those on diabetes medications, *n* = 664.

Bolded values indicate statistically significant (*P* < 0.05) associations.

The interdependent (time displacement) associations with cardio-metabolic biomarkers are shown in Figure 1. Two hours per day increases in stepping time in conjunction with equivalent reductions in standing time were associated with significantly lower BMI ($\approx 10\%$), waist circumference (≈ 7 cm), and 2-h post-load glucose ($\approx 11\%$). Two hours per day increases in stepping time coinciding with equivalent decreases in sitting time were significantly associated with these outcomes ($\approx 11\%$ lower BMI, ≈ 7.5 cm lower waist circumference, and $\approx 12\%$ lower post-load glucose) and with lower triglycerides ($\approx 14\%$) and higher HDL-cholesterol (≈ 0.10 mmol/L). Glucose and lipid profiles were also significantly associated with reallocating time from sitting to standing (posture changes with no additional ambulation). Each 2 h per day sitting-to-standing reallocation was associated with significantly lower fasting glucose ($\approx 2\%$), total/HDL-cholesterol ratio ($\approx 6\%$), triglycerides ($\approx 11\%$), and higher HDL-cholesterol (≈ 0.06 mmol/L). The only inconclusive non-significant findings pertained to associations with waist circumference of sitting-to-standing reallocations and with lipid outcomes of standing-to-stepping reallocations. Further adjustment for stepping intensity mostly did not alter findings (Supplementary material online, Table S7); however, some associations became non-significant. Mostly, this was with little or no changes to effect size except that the standing-to-stepping reallocation had an

association with waist circumference that changed from ≈ 7.0 cm (*P* = 0.001) to ≈ 4.5 cm (*P* = 0.053) upon adjustment.

Discussion

These findings provide novel evidence on associations with cardio-metabolic risk biomarkers of standing, measured objectively from direct postural sensors, in a sample of Australian community-dwelling adults. Importantly, associations were identified with consideration to time displacement (i.e. when sleep is not changed, sitting reductions inevitably require increases to standing time, stepping time, or both). These cross-sectional findings provide some indication that cardio-metabolic benefits, particularly to glucose and lipid metabolism, may be achieved when reducing sitting through increases in standing, at volumes shown to be feasible and acceptable in workplace-setting interventions.^{23,24} Significant associations with indicators of adiposity (BMI and waist circumference) were only observed when additional time was spent stepping (at the expense of either reduced standing or reduced sitting).

A key strength was the use of highly accurate activity monitors that directly measure posture from the thigh position. Prior studies have all relied on self-report, or monitors worn on the hip or waist that are prone to misclassification.²⁵ Findings regarding the

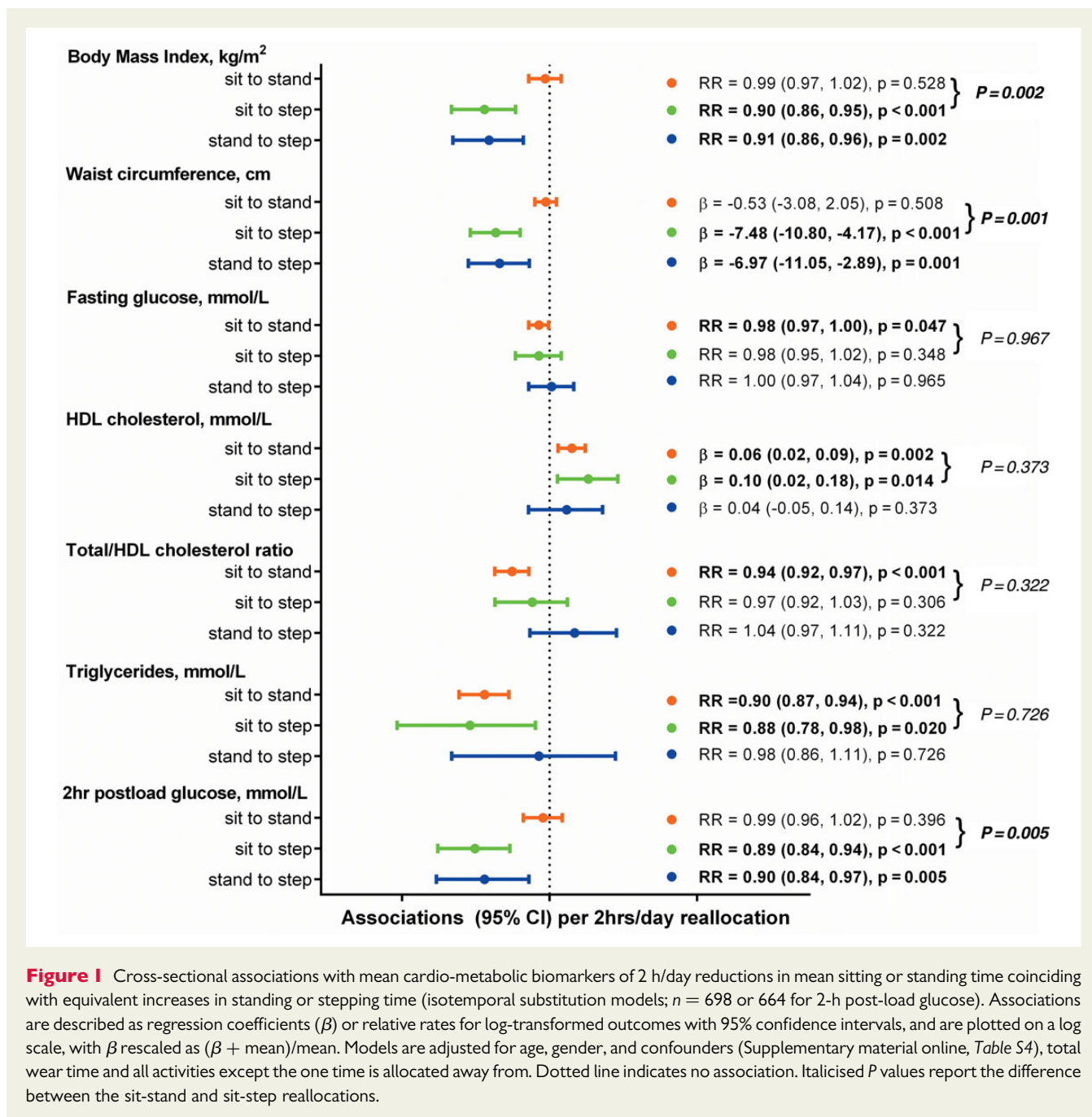


Figure 1 Cross-sectional associations with mean cardio-metabolic biomarkers of 2 h/day reductions in mean sitting or standing time coinciding with equivalent increases in standing or stepping time (isotemporal substitution models; n = 698 or 664 for 2-h post-load glucose). Associations are described as regression coefficients (β) or relative rates for log-transformed outcomes with 95% confidence intervals, and are plotted on a log scale, with β rescaled as $(\beta + \text{mean})/\text{mean}$. Models are adjusted for age, gender, and confounders (Supplementary material online, Table S4), total wear time and all activities except the one time is allocated away from. Dotted line indicates no association. Italicised P values report the difference between the sit-stand and sit-step reallocations.

percentage of the waking day spent sitting/reclining (56%), and the associations of sitting time with the cardio-metabolic biomarkers, were broadly consistent with the extant literature on sedentary time as measured with other methods.^{4,26–28} This is one of the first studies to report on the associations of objectively measured *standing* with cardio-metabolic biomarkers. Standing comprised nearly one-third ($\approx 31\%$) of waking hours and most ($\approx 70\%$) of the waking hours that were not spent sitting. Standing showed beneficial associations with lipids, and fasting and 2-h post-load plasma glucose. The glucose findings are consistent with experimental studies showing acute reductions in postprandial glucose following short bouts of standing,^{9,10} and might be one pathway to explain the lower risk of mortality associated with increased self-reported standing.²⁹ Most associations

of standing with cardio-metabolic outcomes persisted after adjustment for MVPA. However, these ‘independent’ or ‘adjusted’ effects may understate the relevance for each activity to cardio-metabolic health as they exclude any effects due to time displacement mechanisms.

We considered such time displacement effects in the isotemporal substitution analyses. Such approaches have been used before,^{12,13} but with reallocations to light-intensity activity, not specifically between seated and upright posture in the absence of ambulation. The associations with fasting glucose, HDL-cholesterol, and triglycerides were similar whether reallocating time from sitting to standing, or to stepping: findings consistent with some posture-based mechanisms proposed. Specifically, both standing and stepping

increase skeletal muscle activity compared with a sitting or reclining posture.³⁰ In animal models, local muscle contractile activity influences maintenance of lipoprotein lipase activity—one of the key enzymes in glucose and lipid metabolism.³¹ An upright posture is also associated with increased muscle sympathetic nerve activity³² and reductions in plasma volume,³³ which may contribute to the associations observed with the lipid and glucose biomarkers. The association with fasting glucose was modest and of borderline significance ($P = 0.047$); the associations with lipids were more pronounced.

Associations with adiposity biomarkers and post-load plasma glucose appeared to differ depending on the specific activity (standing/stepping) sitting time was reallocated to. Only the reallocations to stepping reached statistical significance. The only inconclusive finding was regarding the association of sitting-to-standing reallocations with waist circumference. These anthropometric findings are consistent with the known importance of energy homeostasis for adiposity,³⁴ and the much greater energy expenditure of stepping compared with sitting or standing³⁵—which was on average in this study ≈ 3.11 METs, 1.40 METs, and 1.25 METs, respectively. Here, 2 h/day sitting reductions would average increases of ≈ 0.30 MET-h when increasing standing, and ≈ 3.72 MET-h when increasing stepping. Though not measured directly, much activity likely occurred outside the exercise context and results therefore may reflect an importance of non-exercise activity thermogenesis.³⁶

The time–displacement findings were broadly consistent with those of previous studies.^{12,13} Specifically, in adults (≥ 20 years) from the US National Health and Nutrition Examination Survey, reallocating time from sedentary to light-intensity activity had significant beneficial associations with triglycerides and markers of insulin resistance, with a reallocation to MVPA necessary to observe significant beneficial associations with waist circumference.¹³ In adults aged 57–79 years from the Whitehall II epidemiological cohort, significant benefits on cardio-metabolic biomarkers were only observed when reallocating time from sedentary to MVPA, with no statistically significant associations observed for the sedentary to light-intensity reallocation.¹²

While experimental studies are required to verify these cross-sectional findings, they nevertheless have relevance for sitting reduction interventions, particularly those that predominantly displace sitting with one alternative activity. With workplace treadmill-desk interventions, sitting is likely to be replaced predominantly with stepping; with workplace sit-stand workstation interventions, sitting reductions are likely to be primarily achieved by increased standing.¹⁴ Accordingly, if both types of intervention approaches achieve a similar sitting reduction, benefits to lipid (and possibly glucose) metabolism are likely to be seen in both interventions, but treadmill-desk interventions (with their associated caveats) may be the approach most likely to achieve the greatest adiposity benefits. To date, the evidence regarding the non-acute (i.e. >1 day) effects of such interventions on biomarkers is mostly inconclusive due to insufficient sample size.³⁷ Nonetheless, some findings support the cross-sectional evidence. Benefits in terms of higher HDL-cholesterol have been reported in an intervention whose participants mostly or exclusively replaced sitting with standing.²⁴ Conversely, benefits for waist circumference and total- and LDL-cholesterol³⁸ and weight³⁹ have been observed in interventions where sitting

was mostly or exclusively replaced with physical activities of a higher intensity (e.g. walking).

Our study involved a moderate sample size that appeared adequately powered for most analyses, except for those instances where associations were non-significant but meaningful effect sizes could not be ruled out. Larger studies are needed for definitive evidence. The sample covered a wide range of community-dwelling adults located across Australia, but loss to follow-up, the subsampling, and participation biases may limit generalizability. Key biomarkers, including insulin and inflammatory markers, that may reflect potential pathways through which sitting and upright activity may impact on cardio-metabolic health,⁴⁰ were not measured. Results may be affected by residual confounding from variables that were not measured or measured with error. For example, with limited monitor removal during waking hours, adjusting for waking wear time indirectly controlled for sleep, but with some residual confounding possible due to non-linearity and measurement error. Residual confounding by stepping intensity is possible in those instances where adjustment led to loss of significance; but this may also have been power loss upon adjustment as the changes to effect sizes were mostly limited. Collinearity problems were incurred when attempting to examine accumulation patterns (prolonged vs. shorter bouts) of sitting and standing that may have different associations with cardio-metabolic biomarkers^{41,42}; accordingly, these were not reported. Longitudinal or intervention studies are required to provide evidence regarding causation.

These findings provide important preliminary evidence on the potential benefits of standing for cardio-metabolic risk biomarkers, especially improved lipid metabolism. This has important public health implications given that standing is a common behaviour,²⁹ the most common alternative to sitting, and predominantly replaces sitting in some types of effective and acceptable environmental sitting-reduction interventions.²⁴ Findings suggest that the potential benefits of sitting reduction is likely to depend on the behaviours with which sitting is replaced; this should be tested by comparing cardio-metabolic outcomes from sitting-reduction trials that achieve comparable sitting reductions by increased standing (e.g. sit-stand workstations and activity-permissive desks) vs. by increased stepping (e.g. treadmill desks).

Supplementary Material

Supplementary Material is available at *European Heart Journal* online.

Acknowledgements

We acknowledge and thank the participants and research staff of AusDiab3.

Funding

This work was supported by National Health and Medical Research Council of Australia (Program Grant #566940 to N.O.; Centre of Research Excellence Grant #1057608 to G.N.H., N.O., and D.D. with salary support to E.W.; Senior Principal Research Fellowship #1003960 to N.O.; Senior Research Fellowship #1078360 to D.W.D.); Heart Foundation Postdoctoral Fellowship (#PH 12B 7054 to G.N.H.); and the Victorian Government's OIS Program (to N.O. and D.W.D.). The AusDiab study co-coordinated by the Baker IDI Heart and Diabetes Institute, gratefully acknowledges the support and

assistance given by: K. Anstey, B. Atkins, B. Balkau, E. Barr, A. Cameron, S. Chadban, M. de Courten, N. Htun, A. Kavanagh, D. Magliano, S. Murray, K. Polkinghorne, J. Shaw, A. Tonkin, T. Welborn, P. Zimmet, and all the study participants. Also, for funding or logistical support, we are grateful to National Health and Medical Research Council (NHMRC grants 233200 and 1007544), Australian Government Department of Health and Ageing, Abbott Australasia Pty Ltd, Alphapharm Pty Ltd, Amgen Australia, AstraZeneca, Bristol-Myers Squibb, City Health Centre-Diabetes Service-Canberra, Department of Health and Community Services—Northern Territory, Department of Health and Human Services—Tasmania, Department of Health—New South Wales, Department of Health—Western Australia, Department of Health—South Australia, Department of Human Services—Victoria, Diabetes Australia, Diabetes Australia Northern Territory, Eli Lilly Australia, Estate of the Late Edward Wilson, GlaxoSmithKline, Jack Brockhoff Foundation, Janssen-Cilag, Kidney Health Australia, Marian & FH Flack Trust, Menzies Research Institute, Merck Sharp & Dohme, Novartis Pharmaceuticals, Novo Nordisk Pharmaceuticals, Pfizer Pty Ltd, Pratt Foundation, Queensland Health, Roche Diagnostics Australia, Royal Prince Alfred Hospital, Sydney, Sanofi Aventis, sanofi-synthelabo, and the Victorian Government's OIS Program. The funders of this study had no role in the data analysis or interpretation of the results.

Conflict of interest: none declared.

References

- Wilmot EG, Edwardson CL, Achana FA, Davies MJ, Gorely T, Gray LJ, Khunti K, Yates T, Biddle SJ. Sedentary time in adults and the association with diabetes, cardiovascular disease and death: systematic review and meta-analysis. *Diabetologia* 2012;**55**:2895–2905.
- Thorp AA, Owen N, Neuhaus M, Dunstan DW. Sedentary behaviors and subsequent health outcomes in adults: a systematic review of longitudinal studies, 1996–2011. *Am J Prev Med* 2011;**41**:207–215.
- Bennie JA, Chau JY, van der Ploeg HP, Stamatakis E, Do A, Bauman A. The prevalence and correlates of sitting in European adults – a comparison of 32 Eurobarometer-participating countries. *Int J Behav Nutr Phys Act* 2013;**10**:107.
- Matthews CE, Chen KY, Freedson PS, Buchowski MS, Beech BM, Pate RR, Troiano RP. Amount of time spent in sedentary behaviors in the United States, 2003–2004. *Am J Epidemiol* 2008;**167**:875–881.
- Colley RC, Garriguet D, Janssen I, Craig CL, Clarke J, Tremblay MS. Physical activity of Canadian adults: accelerometer results from the 2007 to 2009 Canadian Health Measures Survey. *Health Rep* 2011;**22**:7–14.
- Australian Government Department of Health. Australia's physical activity and sedentary behaviour guidelines, Canberra, Australia: Department of Health; 2014.
- Department of Health, Physical Activity, Health Improvement and Protection. Start active, stay active: a report on physical activity from the four home countries' Chief Medical Officers, London, UK: The Department of Health; 2011.
- Physical Activity Guidelines Advisory Committee. Physical Activity Guidelines Advisory Committee Report. Washington, DC: U.S. Department of Health and Human Services, 2008.
- Buckley JP, Mellor DD, Morris M, Joseph F. Standing-based office work shows encouraging signs of attenuating post-prandial glycaemic excursion. *Occup Environ Med* 2014;**71**:109–111.
- Thorp AA, Kingwell BA, Sethi P, Hammond L, Owen N, Dunstan DW. Alternating bouts of sitting and standing attenuates postprandial glucose responses. *Med Sci Sports Exerc* 2014;**46**:2053–2061.
- Mekary RA, Willett WC, Hu FB, Ding EL. Isotemporal substitution paradigm for physical activity epidemiology and weight change. *Am J Epidemiol* 2009;**170**:519–527.
- Hamer M, Stamatakis E, Steptoe A. Effects of substituting sedentary time with physical activity on metabolic risk. *Med Sci Sports Exerc* 2014;**46**:1946–1950.
- Buman MP, Winkler EA, Kurka JM, Hekler EB, Baldwin CM, Owen N, Ainsworth BE, Healy GN, Gardiner PA. Reallocating time to sleep, sedentary behaviors, or active behaviors: associations with cardiovascular disease risk biomarkers, NHANES 2005–2006. *Am J Epidemiol* 2014;**179**:323–334.
- Stephens SK, Winkler EA, Trost SG, Dunstan DW, Eakin EG, Chastin SF, Healy GN. Intervening to reduce workplace sitting time: how and when do changes to sitting time occur? *Br J Sports Med* 2014;**48**:1037–1042.
- Tanamas SK, Magliano DJ, Lynch BM, Sethi P, Willenberg L, Polkinghorne KR, Chadban S, Dunstan DW, Shaw JE. *AusDiab* 2012. The Australian Diabetes, Obesity and Lifestyle Study. Melbourne, VIC, Australia: Baker IDI Heart and Diabetes Institute, 2013.
- Dunstan DW, Zimmet PZ, Welborn TA, Cameron AJ, Shaw J, de Courten M, Jolley D, McCarty DJ, Australian Diabetes O, Lifestyle S. The Australian Diabetes, Obesity and Lifestyle Study (AusDiab) – methods and response rates. *Diabetes Res Clin Pract* 2002;**57**:119–129.
- Kozey-Keadle S, Libertine A, Lyden K, Staudenmayer J, Freedson PS. Validation of wearable monitors for assessing sedentary behavior. *Med Sci Sports Exerc* 2011;**43**:1561–1567.
- Grant PM, Ryan CG, Tigbe WW, Granat MH. The validation of a novel activity monitor in the measurement of posture and motion during everyday activities. *Br J Sports Med* 2006;**40**:992–997.
- Ryan CG, Grant PM, Tigbe WW, Granat MH. The validity and reliability of a novel activity monitor as a measure of walking. *Br J Sports Med* 2006;**40**:779–784.
- Lyden K, Kozey Keadle SL, Staudenmayer JW, Freedson PS. Validity of two wearable monitors to estimate breaks from sedentary time. *Med Sci Sports Exerc* 2012;**44**:2243–2252.
- Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem* 1972;**18**:499–502.
- Giles GG, Ireland PD. *Dietary questionnaire for epidemiological studies (Version 2)*. Melbourne: Cancer Council Victoria, 1996.
- Healy GN, Eakin EG, Lamontagne AD, Owen N, Winkler EA, Wiesner G, Gunning L, Neuhaus M, Lawler S, Fjeldsoe BS, Dunstan DW. Reducing sitting time in office workers: short-term efficacy of a multicomponent intervention. *Prev Med* 2013;**57**:43–48.
- Alkhajah TA, Reeves MM, Eakin EG, Winkler EA, Owen N, Healy GN. Sit-stand workstations: a pilot intervention to reduce office sitting time. *Am J Prev Med* 2012;**43**:298–303.
- Aguilar-Farías N, Brown WJ, Peeters GM. ActiGraph GT3X+ cut-points for identifying sedentary behaviour in older adults in free-living environments. *J Sci Med Sport* 2014;**17**:293–299.
- Henson J, Yates T, Biddle SJ, Edwardson CL, Khunti K, Wilmot EG, Gray LJ, Gorely T, Nimmo MA, Davies MJ. Associations of objectively measured sedentary behaviour and physical activity with markers of cardiometabolic health. *Diabetologia* 2013;**56**:1012–1020.
- Healy GN, Matthews CE, Dunstan DW, Winkler EA, Owen N. Sedentary time and cardio-metabolic biomarkers in US adults: NHANES 2003–06. *Eur Heart J* 2011;**32**:590–597.
- Hagstromer M, Troiano RP, Sjostrom M, Berrigan D. Levels and patterns of objectively assessed physical activity – a comparison between Sweden and the United States. *Am J Epidemiol* 2010;**171**:1055–1064.
- Katzmarzyk PT. Standing and mortality in a prospective cohort of Canadian adults. *Med Sci Sports Exerc* 2014;**46**:940–946.
- Tikkanen O, Haakana P, Pesola AJ, Hakkinen K, Rantalainen T, Havu M, Pullinen T, Finni T. Muscle activity and inactivity periods during normal daily life. *PLoS ONE* 2013;**8**:e52228.
- Hamilton MT, Hamilton DG, Zderic TW. Exercise physiology versus inactivity physiology: an essential concept for understanding lipoprotein lipase regulation. *Exerc Sport Sci Rev* 2004;**32**:161–166.
- Convertino VA, Cooke WH. Relationship between stroke volume and sympathetic nerve activity: new insights about autonomic mechanisms of syncope. *J Gravit Physiol* 2002;**9**:P63–P66.
- Jacob G, Ertl AC, Shannon JR, Furlan R, Robertson RM, Robertson D. Effect of standing on neurohumoral responses and plasma volume in healthy subjects. *J Appl Physiol* 1998;**84**:914–921.
- Hunter GR, Byrne NM. Physical activity and muscle function but not resting energy expenditure impact on weight gain. *J Strength Cond Res* 2005;**19**:225–230.
- Levine JA, Schleusner SJ, Jensen MD. Energy expenditure of nonexercise activity. *Am J Clin Nutr* 2000;**72**:1451–1454.
- Levine JA. Non-exercise activity thermogenesis (NEAT). *Best Pract Res Clin Endocrinol Metab* 2002;**16**:679–702.
- Neuhaus M, Eakin EG, Straker L, Owen N, Dunstan DW, Reid N, Healy GN. Reducing occupational sedentary time: a systematic review and meta-analysis of evidence on activity-permissive workstations. *Obes Rev* 2014;**15**:822–838.
- John D, Thompson DL, Raynor H, Bielak K, Rider B, Bassett DR. Treadmill workstations: a worksite physical activity intervention in overweight and obese office workers. *J Phys Act Health* 2011;**8**:1034–1043.
- Koepp GA, Manohar CU, McCrady-Spitzer SK, Ben-Ner A, Hamann DJ, Runge CF, Levine JA. Treadmill desks: a 1-year prospective trial. *Obesity* 2013;**21**:705–711.
- Hamilton MT, Hamilton DG, Zderic TW. Role of low energy expenditure and sitting in obesity, metabolic syndrome, type 2 diabetes, and cardiovascular disease. *Diabetes* 2007;**56**:2655–2667.
- McCulloch J. Health risks associated with prolonged standing. *Work* 2002;**19**:201–205.
- Carson V, Wong SL, Winkler E, Healy GN, Colley RC, Tremblay MS. Patterns of sedentary time and cardiometabolic risk among Canadian adults. *Prev Med* 2014;**65**:23–27.