

**This is an electronic reprint of the original article.
This reprint *may differ* from the original in pagination and typographic detail.**

Author(s): Gao, Ying; Silvennoinen, Mika; Pesola, Arto; Kainulainen, Heikki; Cronin, Neil; Finni Juutinen, Taija

Title: Acute Metabolic Response, Energy Expenditure, and EMG Activity in Sitting and Standing

Year: 2017

Version:

Please cite the original version:

Gao, Y., Silvennoinen, M., Pesola, A., Kainulainen, H., Cronin, N., & Finni Juutinen, T. (2017). Acute Metabolic Response, Energy Expenditure, and EMG Activity in Sitting and Standing. *Medicine and Science in Sports and Exercise*, 49(9), 1927-1934.
<https://doi.org/10.1249/MSS.0000000000001305>

All material supplied via JYX is protected by copyright and other intellectual property rights, and duplication or sale of all or part of any of the repository collections is not permitted, except that material may be duplicated by you for your research use or educational purposes in electronic or print form. You must obtain permission for any other use. Electronic or print copies may not be offered, whether for sale or otherwise to anyone who is not an authorised user.

Medicine & Science IN Sports & Exercise

The Official Journal of the American College of Sports Medicine
www.acsm-msse.org

. . . Published ahead of Print

Acute Metabolic Response, Energy Expenditure, and EMG Activity in Sitting and Standing

Ying Gao, Mika Silvennoinen, Arto Pesola, Heikki Kainulainen, Neil J. Cronin, and Taija Finni

Neuromuscular Research Center, Biology of Physical Activity, Faculty of Sport and Health
Sciences, University of Jyväskylä, Finland

Accepted for Publication: 13 April 2017

Medicine & Science in Sports & Exercise, **Published ahead of Print** contains articles in unedited manuscript form that have been peer reviewed and accepted for publication. This manuscript will undergo copyediting, page composition, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered that could affect the content.

Copyright © 2017 American College of Sports Medicine

Acute Metabolic Response, Energy Expenditure, and EMG Activity in Sitting and Standing

Ying Gao, Mika Silvennoinen, Arto Pesola, Heikki Kainulainen, Neil J. Cronin, and Taija Finni

Neuromuscular Research Center, Biology of Physical Activity, Faculty of Sport and Health
Sciences, University of Jyväskylä, Finland

Corresponding author information:

Ying Gao

Neuromuscular Research Center

Department of Biology of Physical Activity

University of Jyväskylä

P.O. Box 35

FI-40014 University of Jyväskylä

Finland

Tel. +358 40 029 1968

Fax. +358 14 260 2071

E-mail. yigao@jyu.fi

Ying Gao was supported by the China Scholarship Council (NO.201206320092), China. The authors declare that they have no competing interests.

ABSTRACT

Purpose: While merely standing up interrupts sedentary behavior, it is important to study acute metabolic responses during single bouts of sitting and standing to understand the physiological processes affecting the health of office workers. **Methods:** 18 healthy middle aged women aged 49.4 ± 7.9 years (range: 40 to 64) with a BMI of $23.4 \pm 2.8 \text{ kg}\cdot\text{m}^{-2}$ volunteered for this laboratory-based randomized crossover trial where they performed two hours desk work either in sitting or standing postures after overnight fasting. Muscle activity (normalized to walking at 5 km/h), respiratory gas exchange and blood samples were assessed following glucose loading (75 g). **Results:** Compared with seated work, continuous standing resulted in greater activity in the thigh muscles (mean of biceps femoris and vastus lateralis: $17 \pm 8\%$ vs. $7 \pm 2\%$, $p < 0.001$), and leg muscles (mean of tibialis anterior, gastrocnemius medialis and soleus: $16 \pm 6\%$ vs. $7 \pm 3\%$, $p < 0.001$), but no increases in back muscle activity (thoracic erector spinae, lumbar erector spinae and multifidus). Concomitant with ~9% higher energy expenditure (EE) ($p = 0.002$), standing resulted in higher fat oxidation ($48 \pm 9\%$ EE vs. $39 \pm 7\%$ EE, $p = 0.008$) and lower carbohydrate oxidation ($52 \pm 9\%$ EE vs. $61 \pm 7\%$ EE, $p = 0.008$) than sitting. Glucose total and net incremental area under the curve were ~10% ($p = 0.026$) and ~42% ($p = 0.017$) higher during standing than sitting, respectively. Insulin concentration did not differ between conditions. **Conclusion:** Compared to sitting, two hours of standing increased muscle activity, fat oxidation and circulating glucose level. These results suggest fuel switching in favor of fat oxidation during standing despite extra carbohydrate availability.

KEYWORDS: Carbohydrate oxidation, energy expenditure, fat oxidation, glucose loading, muscle activity, sit-stand workstation

INTRODUCTION

Sedentary behavior is defined as a sitting or reclining posture with low energy expenditure (≤ 1.5 METs) (28). Sedentary behavior is associated with increased risks of type 2 diabetes, cardiovascular disease, and cardiovascular and all-cause mortality (5). At the population level, the amount of moderate-to-vigorous physical activity is insufficient to offset the health risks of a high amount of sedentary time (13). Standing is linked to lower all-cause and cardiovascular disease mortality (20, 31), suggesting that reducing sedentary time by standing may be beneficial. However, it is still unclear if and why standing might be a healthy substitute for sitting (15).

Standing instead of sitting has been shown to decrease acute postprandial glucose responses without affecting insulin responses (30). This may be due to improved contraction-mediated glucose uptake (3) as a result of increased muscle activity during standing (24). Moreover, increased fat oxidation during physical activity might improve glucose tolerance indirectly through improved muscle lipid uptake, trafficking and oxidation, which serves to clear insulin-inhibiting fat metabolites within muscle cells (4). Therefore, standing may benefit glucose tolerance through mechanisms linked to either increased carbohydrate *or* fat oxidation, but these mutually inhibitory mechanisms have not been quantified concurrently with muscle activity and metabolic markers during standing. Concurrent measurement of these potential mechanisms is required to explain why in some studies standing has not elicited metabolic benefits (1, 23), and thus to elucidate whether standing is a healthy alternative to sitting.

The purpose of this study was to investigate acute physiological responses to two hours of sitting and standing work postures, including muscle activity, energy expenditure, fat and carbohydrate oxidation, glucose tolerance and insulin response after glucose loading. The main hypotheses of this study were that compared to sitting, continued standing at work following glucose loading would 1) increase energy expenditure through greater muscle activity in the lower limbs, 2) reduce glucose responses without effects on insulin responses, and 3) increase fat oxidation despite the glucose loading.

METHODS

Recruitment and study sample

This study was carried out at the University of Jyväskylä, Finland, in the Faculty of Health and Sport Sciences. Ethical approval for the study was granted by the Ethics Committee of the University of Jyväskylä (27/3/2015). Recruitment was performed in the Jyväskylä region by posting advertisements to the University of Jyväskylä website and public places. Individuals who were interested in the study were contacted by email. Inclusion criteria were: healthy female, age range from 40 to 65 years old with a heightened diabetes risk (10), non-pregnant, non-smoker, able to perform two hours of continuous sitting and standing, and a desk-based occupation involving sedentary tasks. Exclusion criteria were: self-reported chronic, long-term musculoskeletal disease, clinically diagnosed diabetes, and cardiovascular or metabolic disease requiring medication known to affect metabolism. Subjects were individually face-to-face informed about the procedures, risks and benefits of the study, and they signed a written informed consent before any measurements. All subjects were volunteers with the right to

withdraw from the study at any time without specifying a reason and without consequences. No monetary incentive was offered to the subjects.

Sample size calculations were based on our pilot data ($n = 6$) of mean changes in the total area under the curve (tAUC) and the net incremental area under the curve (iAUC) for plasma glucose (88 mmol/L·min, SD 134 mmol/L·min and 72 mmol/L·min, SD 105 mmol/L·min, respectively). A sample size of 18 was assumed to provide at least 80% power (5% significance, two-tailed) to detect glucose differences within subjects between sitting and standing. This would also have sufficient power (at least 90%) to detect differences in the other main outcomes: energy expenditure (relative difference of $11 \pm 9\%$) and muscle activity of quadriceps and hamstring muscles (relative difference of $78 \pm 110\%$).

Study design and protocol

In this randomized crossover controlled study, subjects performed 2 h desk work either in sitting or standing on separate days starting at the same time of day. The order of sit/stand conditions was randomized using online software (www.randomizer.org). We used a minimum wash-out of six days (the maximum was 21 days) between measurement days to eliminate any potential carryover effects. At the beginning of the study, subjects were invited to attend a brief familiarization session. At the familiarization session, questionnaires about background information were completed and subjects were familiarized with the office setting of the laboratory, which included basic office equipment (computer and internet) and an electric, height-adjustable sit-stand workstation (ISKU, Finland). The height of the sit-stand workstation was individually adjusted for sitting or standing, and during the sitting task, the height of an

office chair was individually adjusted according to ergonomic recommendations (Finnish Institute of Occupational Health: <http://www.ttl.fi/en/Pages/default.aspx>).

For two days before the measurement day, subjects were asked to wear a triaxial accelerometer (X6-1a, Gulf Coast Data Concepts Inc, USA) on the right side of the waist to monitor physical activity during waking hours, and to keep a log of wear/nonwear time and sleep time. Within these two days subjects were asked to refrain from any exercise training and alcohol, and from caffeine at least 12 hours before the measurement day. For one day before the measurements, subjects filled in a detailed diet diary including time of meals and volume and type of food and drinks consumed. Subjects were also requested to obey the same diet the day before the second measurement day. All subjects were provided with verbal and written instructions.

The timeline of the measurement day is shown in Figure 1. On each measurement day, subjects were instructed to minimize physical activity and to drive or take the bus to the research laboratory in the morning at 08:00, after a 12-h overnight fasting. Baseline assessments included anthropometry and body composition. Electrodes for measuring muscle activity were attached to eight muscles. A heart rate monitor and individually fitted gas exchange mask were attached. After all equipment were in place and set to record, subjects sat quietly in the initial preparatory phase for 45 minutes. Fasting venous blood samples were then taken whilst seated (time point 0 min). Immediately afterwards they were given a standard oral glucose loading (250 ml glucose drink with 75 g of glucose) containing 450 KJ (110 kcal) of energy (GlucosePro, COMED, Tampere, Finland). The workstation was then individually adjusted for sitting or standing and the subject began computer work or reading a book for the next two hours. The same chosen task

was performed on both measurement days. During the standing condition the subjects were allowed to sway and bend their legs but movement was otherwise restricted due to the position of research devices. Venous blood samples were retaken at 30, 60, and 120 min. At the end of the measurement, with the bipolar electrodes still on, the subjects were asked to walk on a treadmill (OJK-1, Telineyhtymä, Kotka, Finland) at 5 km/h for one minute.

Assessments and analysis

Demographics, anthropometry and body composition. The background questionnaires included socio-demographic, work-related and health-related items. Physical fitness was assessed with a non-exercise questionnaire (NASA/JSC Physical Activity Scale during the last month; PA-R-1m) (27). Subjects were weighed in a fasted state using the same digital scale wearing minimal clothes and without shoes. On one of the measurement mornings, subjects' height and body composition (InBody 720, Biospace Ltd, Seoul, Korea) were measured in a fasted condition yielding body mass, skeletal muscle mass, body free fat mass, body fat mass, percent body fat and body mass index (BMI).

Physical activity, sleep time and diet recordings. A triaxial accelerometer was used to monitor physical activity during waking time, except water based activities. Data were recorded in 1-min epochs with accelerometer counts less than 100 counts/min classified as sedentary time, 101–1952 counts/min as light-intensity activity, and more than 1952 counts/min as moderate-to-vigorous intensity activity (14). Wear time and non-wear periods were determined from daily logs. Based on self-reports, sleep time was determined in minutes. Dietary records were analyzed

using web-based dietary recall (Nutri-Flow Oy, 2015, <http://nutri-flow.fi/>, Finland) to determine energy intake and macronutrient content including fat, protein and carbohydrate.

Venous blood samples were collected and analyzed using standardized clinical procedures for serum lipids and glycerol, plasma glucose (Konelab 20 Xti, Thermo Fisher Scientific Oy, Vantaa, Finland), and serum insulin and cortisol (Immulite 2000 Xpi, Siemens Healthcare Diagnostics., United Kingdom). The intra-assay coefficients of variation were 2.4% for triglycerides, 1.7% for glycerol, 2.8% for fat free acids (FFA), 7.8% for cortisol, 1.7% for glucose, and 4.2% for insulin.

From fasting blood (0 min), total cholesterol, high- (HDL-C) and low-density lipoprotein cholesterol (LDL-C), triglycerides, glycerol, FFA, cortisol, glucose and insulin were analyzed. After glucose loading, the blood samples were analyzed for triglycerides, glycerol, FFA, cortisol, glucose and insulin at several time points (30, 60 and 120 min). The few missing samples (n = 1 at 30 min and n = 1 at 60 min) were interpolated using the best fit of a second degree polynomial through the other sample points available from the same individual. The total area under the curve (tAUC) and the net incremental area under the curve (iAUC) of a 120 minute period were calculated for glucose, insulin, triglycerides, glycerol, FFA and cortisol using a trapezoidal approximation of area under the curve, where tAUC was calculated from the zero level and iAUC from the fasting level.

Indirect calorimetry and heart rate. Subjects breathed through a facial mask equipped with ventilation sensors and gas sampling tubes. Ventilation, oxygen consumption (VO_2) and carbon dioxide production (VCO_2) were measured (breath by breath) with a Jaeger Oxycon Pro and

LabManager 3.0 software (Viasys Healthcare GmbH, Hoechberg, Germany). The measurement system was calibrated before each measurement and standardized for barometric pressure, temperature and humidity. Outputs of ventilation (VE), breathing frequency (BF), VO_2 , VCO_2 , VO_2/kg , respiratory exchange ratio (RER) and absolute metabolic equivalent (MET) were collected and averaged over 30 s intervals for data analysis. Energy expenditure and the percentage of fat and carbohydrate usage for energy production were calculated using respiratory quotient values, with corresponding caloric equivalent values (without protein) and oxygen uptake. For the preparatory phase, a moving average was analyzed over 15 minute periods. The lowest values were taken to represent a steady state, where the mean resting energy expenditure was 0.9 ± 0.1 kcal/min, and ratios of fat and carbohydrate energy were $60.0 \pm 10.8\%$ EE and $40.0 \pm 10.8\%$ EE, respectively. During sitting and standing work the periods where the mask was removed were discarded and the mean values of both 2 h conditions were calculated for the main variables.

Heart rate (HR) was measured using a heart rate belt (Polar Electro Oy, Finland) with a Polar RS800CXtm wrist computer. HR was recorded every 5 s for the duration of the measurement and averaged over the 2 h measurement period.

Electromyography. Surface electromyography (EMG) was used to study the activity of back and lower limb muscles throughout the measurements. Standard electrode placement and skin preparation procedures were used (18). Bipolar electrodes (Ag/AgCl, Ambu White Sensor, 4500M, USA) were attached unilaterally on the right side over the following muscles: thoracic erector spinae (TES), lumbar erector spinae (LES), lumbar multifidus (LM), biceps femoris (BF),

vastus lateralis (VL), tibialis anterior (TA), gastrocnemius medialis (GM) and soleus (SOL), all with an inter-electrode distance of 20 mm. EMG amplitude was normalized channel by channel and expressed as a percentage of that during walking at 5 km/h on a treadmill (%walk). The signals were collected using ME6000 Biomonitor, and root mean square (RMS) values from the raw EMG data were computed with Megawin software (Mega Electronics Ltd, Kuopio, Finland). In order to reflect the overall muscle activity level, normalized data from different muscles were averaged to produce mean overall muscle activity. In addition, mean back muscle activity of TES, LES and LM, mean thigh muscle activity of BF and VL, and mean leg muscle activity of TA, GM and SOL were calculated.

Statistical analysis

Statistical analyses were conducted using IBM SPSS for Windows 22.0 (SPSS Inc., Chicago, IL, USA). Values are reported as means \pm standard deviations or % (number) unless otherwise indicated. Tests of normality (Shapiro-Wilk) were applied. Paired t-tests (normal data) or Wilcoxon signed rank tests (non-normal data) were used to assess differences in baseline assessment variables including body mass, dietary parameters and physical activity, and fasting variables between measurement days. For the condition effects of sitting and standing, paired t-tests or Wilcoxon signed rank tests were used to evaluate metabolic markers, mean energy expenditure, mean muscle activity level and mean HR within subjects. Spearman's correlation coefficient (r) was used to assess the strength of correlations between muscle activity and potential parameters including metabolic responses and energy expenditure, respectively. A probability level of $p < 0.05$ (two-tailed) was considered statistically significant.

RESULTS

Sample description and characteristics

Of the 29 subjects who met the inclusion criteria and were interviewed, six were excluded due to medications and five withdrew due to scheduling difficulties. Finally, 18 healthy females were included in the study. Their age ranged between 40 – 64 years. 12 were post-menopausal and six were peri-menopausal. For peri-menopausal females, the menstrual cycle phase was not determined but the measurements were not done during menstruation. The possible influence of menstrual cycle status on the results was tested in a separate analysis with an independent t-test and was found not to influence the results (data not shown). Two subjects stopped the measurements after the first hour in standing work because of feeling faint and unwell, leaving a total of 16 subjects who completed both conditions. From the two subjects with full data during sitting, but with incomplete data during standing, we analyzed their first hour of data regarding mean energy expenditure, muscle activity and HR. The characteristics of all subjects are presented in Table 1. There were no differences in baseline assessments (anthropometric, dietary, and physical activity measures) or fasting biochemical values measured at 0 min between measurement days (Table 2), nor were there significant differences in resting energy expenditure or normalized EMG activities during either of the preparatory phases.

Muscle activity

The effects of condition on EMG activity are presented in Table 3. The muscle groups were categorized and averaged by region, where back muscles included TES, LES and LM, thigh muscles included BF and VL, and leg muscles included TA, GM and SOL. During continued standing, the overall muscle activity level of the back, thigh and leg muscles combined was

49.4% greater than during sitting ($26.4 \pm 9.4\%$ vs. $19.1 \pm 5.9\%$, $p = 0.006$). This difference resulted from 173.6% greater activity in thigh muscles ($17.2 \pm 8.4\%$ vs. $6.9 \pm 2.1\%$, $p < 0.001$) and 160.5% greater activity in leg muscles ($15.9 \pm 6.1\%$ vs. $7.0 \pm 2.5\%$, $p < 0.001$), but no significant differences in the activity of back muscles ($39.0 \pm 16.6\%$ vs. $43.0 \pm 18.4\%$, $p > 0.05$). Detailed results from different muscle groups are presented in Supplemental Digital Content 1 (see Figure, Supplemental Digital Content 1, muscle activity of different muscle groups, <http://links.lww.com/MSS/A929>).

Energy expenditure

The results of energy expenditure and HR between conditions can be found in Table 3. Compared to sitting, during two hours of standing desk work the mean of total energy expenditure was 9.2% greater ($p = 0.002$) and the proportion of fat use increased from 39.4% to 48.3%EE ($p = 0.008$) while the proportion of carbohydrate use decreased from 60.6% to 51.7%EE ($p = 0.008$). Concomitant with energy expenditure, standing work resulted in 12.0% higher HR than sitting ($p < 0.001$). Energy expenditure positively correlated with mean thigh ($r = 0.392$, $p = 0.022$) and leg muscle activity ($r = 0.378$, $p = 0.028$).

Metabolic markers

Figure 2 and Table 3 show the metabolic responses to glucose loading in sitting and standing conditions. A significantly higher tAUC (9.8%, $p = 0.026$) and net iAUC of plasma glucose (42.3%, $p = 0.017$) were measured during standing than sitting. After glucose loading, the mean concentration of plasma glucose continued to rise until 60 min, reaching 9.3 ± 2.6 mmol/l during standing, whereas for seated work glucose peaked at 8.6 ± 1.2 mmol/l at the 30 min time point.

For tAUC, net iAUC and changes in 2-h concentration levels of serum insulin, triglycerides, glycerol, FFA and cortisol were not significantly different between conditions. There were no significant correlations between any of the metabolic responses and muscle activity.

DISCUSSION

The present study provides experimental evidence for the effects of two hour bouts of sitting and standing postures on acute metabolic responses, energy expenditure and muscle activity after glucose loading in middle aged women. In line with the hypothesis, standing resulted in greater muscle activity, higher energy expenditure and fat oxidation when compared with sitting. In contrast to our hypothesis, standing elicited a higher glucose response after glucose loading than was observed during sitting. Together these results suggest fuel switching after glucose loading, whereby fat oxidation increased and carbohydrate usage decreased during standing compared to sitting (Figure 3).

Previous studies using indirect calorimetry have reported that continuous motionless standing consumes 0.07 kcal/min more energy than sitting (19). In the present study the energy expenditure increase from sitting to standing was roughly similar (0.10 kcal/min), suggesting that the subjects were mainly standing still during the experiment, despite being allowed to sway and bend their legs. Lower extremity muscle activity was positively associated with energy expenditure, confirming that lower extremity muscle activity is an important factor in energy expenditure during standing. However, neither muscle activity nor energy expenditure were associated with metabolic changes, suggesting that factors other than lower extremity muscle

activity or total energy expenditure may explain how individuals gain acute metabolic benefits when standing still instead of sitting.

In the present study we found an increase in fat oxidation and a decrease in carbohydrate oxidation in standing compared to sitting. This indicates a proportional increase in the use of fatty acids as an energy source and enhanced fatty acid oxidation to fuel muscle activity, which in turn supports the hypothesis that light intensity physical activity like standing may alter the regulation of fat and carbohydrate usage (29). In the long term, increased fat oxidation may help in the clearance of insulin-inhibiting fat metabolites and ectopic fat storage, with beneficial effects on the whole body, as well as muscle and liver insulin sensitivity, even in the absence of a negative energy balance or acute improvements in insulin sensitivity (3, 4). Although in this study glucose loading caused no difference in insulin response or changes in triglyceride levels between conditions at 2 h, our results corroborate those of earlier studies which reported that postprandial insulinemic and lipaemia responses did not significantly change after alternating bouts of standing and sitting for 30 to 45 minutes (23, 30). Romijn et al. (1993) showed that during light intensity exercise, FFA release from adipose tissue is the main oxidative fuel used by working muscles, and lipolysis increases as a function of power output when changing from rest to physical activity (26). It is also probable that increased muscle activity during standing increased FFA delivery into the muscle via increased blood flow (29). While a similar mobilization of FFA took place in both conditions, a slightly slower decline of FFA concentration can be found during standing than sitting (17), although this effect was nonsignificant in the present study. Thus, we speculate that standing may attenuate insulin-induced lipolysis inhibition because glycerol and FFA both tended to decrease slower than

during sitting. The second major source of fat is the release of FFA from triglycerides stored directly in the muscle, which increases during light to moderate intensity exercise (29). Therefore, we conclude that in the conditions of the present study where there was maximal availability of glucose, the increased energy demand during standing promoted fuel switching by increasing fat oxidation (potentially due to increased delivery of FFA and/or increased oxidation of intramuscular FFA) and decreasing carbohydrate oxidation.

In the current study, the elevated level of circulating plasma glucose found in standing suggests that glucose may not be needed as an extra energy source in standing. This apparently conflicts with previous results showing attenuated blood glucose excursion in standing (6, 30). However, previous studies have used standardized standing breaks (30) or standing while working in a real office environment (6) as their exposure, both of which may elicit higher energy expenditure due to dynamic activity compared to predominantly motionless standing in the present study. Buckley et al. (2014) reported that the increase in energy expenditure of standing vs sitting while doing office work was 0.83 kcal/min (6), which is higher than in the present study (0.10 kcal/min). This suggests that frequent standing breaks or ambulation may be required to elevate energy expenditure above that of motionless standing in order to elicit changes in glucose tolerance. Furthermore, the high energy expenditure in Buckley's study was estimated from HR rather than using indirect calorimetry, which may also explain the differences between Buckley's and our findings (6). Many other factors may contribute to the apparent discrepancy regarding the glycemic response, including age, sex, BMI, metabolism and exercise status (6, 30). For example, benefits of standing may be more evident in subjects with a higher BMI than those in the present study (30), since higher muscle activity has been reported in overweight compared to

normal weight subjects (24). Another important difference to consider when comparing results is the time course of nutrient loading. In the present study the loading was done during the standing exposure, because we aimed to study concurrent interaction between standing and nutrient loading, as occurs during daily life, where periods of energy intake and expenditure take place simultaneously. Some setups provide the nutrient loading after the physical activity exposure and do not allow direct comparison to the present findings because of a lack of concurrent interaction between diet and physical activity (11, 30). There is discrepancy in the literature regarding differences in experimental design that may be the cause of these inconsistent findings, e.g. intensity and frequency of breaks and duration of prolonged sitting (1, 9, 16, 17, 30). Importantly, distinct from the majority of experimental studies which have interrupted sitting with short periods of activities (2), the current setup differentiates the independent effects of sitting and standing. It should be noted that although the increased glucose level may seem to induce an adverse effect, the increased oxidation of lipids can benefit insulin sensitivity after intervention or in the long term, and the resulting effect may be positive (4, 9). However, this should be confirmed in longitudinal studies.

Methodological considerations

In this laboratory study, we used a controlled measurement environment in order to eliminate potential confounding factors. In order to simulate a normal office work environment as closely as possible, subjects were first familiarized with the laboratory layout, and during the measurements we asked them to perform their usual daily tasks, which included Internet browsing, emailing, word document editing, reading materials and other paper work. Furthermore, subjects were asked to do the same task during both experimental days in order to

have comparable conditions. There were no differences between conditions at the baseline assessment, suggesting that the changes observed were due to changes in posture as opposed to external factors. However, some between-subjects variance in dietary patterns and profiles of fat, protein and carbohydrate may influence the results. Future studies should standardize meals prior to measurement days to minimize possible dietary effects on responsiveness. Moreover, previous studies have provided a non-standardized lunch or a mixed test drink rather than a glucose drink during the experiment (6, 30), which may induce different changes in postprandial blood glucose responses due to the higher intake of energy and other macronutrients. Previous evidence suggests that differences in nutritional composition can influence plasma glucose concentrations, whereby postprandial plasma glucose concentration was significantly higher in a group that consumed a glucose drink than a group that consumed a drink with glucose and protein (25).

It is important to note that the acute effects observed after two hours of exposure to continuous sitting and standing may not be extrapolated to long-term exposures. The current setup also limited ambulatory activity due to the measurements of respiratory gases and EMG. Unilateral muscle activity may have caused loss of some information about postural variations during the measurements. Furthermore, this study was designed to include single bouts of two hours continuous sitting/standing, with the goal of inducing explicit physiological changes under standardized conditions. It should be noted that a period of two hours continuous standing may not be suitable for all subjects. We were not able to measure full data from two subjects during standing work, as they reported feeling faint and unwell after the first hour. This should be carefully considered in future studies, as ergonomic recommendations suggest that continuous

standing should be limited to one hour, and include frequent adjustments of posture throughout the workday (8). Furthermore, before suggesting the potential effects of promoting standing instead of sitting, a number of health- and work-related outcomes should be considered such as lower limb discomfort and fatigue (7), entire body tiredness, alertness and performance (12), leg swelling and venous blood pooling (21), and low back pain (22). Future studies should also aim to identify the positive and negative effects of sitting/standing during desk work, not only in a lab setting but also in an ecological environment.

CONCLUSION

Maintaining a standing posture increased muscle activity, energy expenditure and plasma glucose concentration compared to sitting following a glucose loading. Standing seems to induce fuel switching in favor of fat oxidation for energy production, which may originate either from oxidation of local fat stores or from elsewhere via delivery in the bloodstream.

ACKNOWLEDGEMENTS AND CONFLICT OF INTEREST

Ying Gao was supported by the China Scholarship Council (NO.201206320092), China. The Center for Scientific Computing (Espoo, Finland) is acknowledged for computer resources. The authors declare that they have no competing interests. The results of the present study do not constitute endorsement by the American College of Sports Medicine. We declare that the results of the study are presented clearly, honestly, and without fabrication, falsification, or inappropriate data manipulation.

REFERENCES

1. Bailey DP, Locke CD. Breaking up prolonged sitting with light-intensity walking improves postprandial glycemia, but breaking up sitting with standing does not. *J Sci Med Sport* 2015;18(3):294–8.
2. Benatti FB, Ried-Larsen M. The Effects of Breaking up Prolonged Sitting Time: A Review of Experimental Studies. *Med Sci Sport Exerc* 2015;47(10):2053–61.
3. Bergouignan A, Latouche C, Heywood S, et al. Frequent interruptions of sedentary time modulates contraction- and insulin-stimulated glucose uptake pathways in muscle: Ancillary analysis from randomized clinical trials. *Sci Rep* 2016;6:32044.
4. Bergouignan A, Momken I, Lefai E, et al. Activity energy expenditure is a major determinant of dietary fat oxidation and trafficking, but the deleterious effect of detraining is more marked than the beneficial effect of training at current recommendations. *Am J Clin Nutr* 2013;98(3):648–58.
5. Biswas A, Oh PI, Faulkner GE, et al. Sedentary Time and Its Association With Risk for Disease Incidence, Mortality, and Hospitalization in Adults. *Ann Intern Med* 2015;162(2):123–32.
6. 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(2):109–11.
7. Chester MR, Rys MJ, Konz SA. Leg swelling, comfort and fatigue when sitting, standing, and sit/standing. *Int J Ind Ergon* 2002;29(5):289–96.
8. Commissaris DACM, Douwes M, Schoenmaker N, de Korte EM. Recommendations for

sufficient physical activity at work. Oxford (UK); [cited 2017 April 17]. Available from:
[http://tools.nisb.nl/beleidsinstrumenten/bravo-](http://tools.nisb.nl/beleidsinstrumenten/bravo-kompas/bewegen/tno_kv1_richtlijnen_voldoende_bewegen_op_werk.pdf)

[kompas/bewegen/tno_kv1_richtlijnen_voldoende_bewegen_op_werk.pdf](http://tools.nisb.nl/beleidsinstrumenten/bravo-kompas/bewegen/tno_kv1_richtlijnen_voldoende_bewegen_op_werk.pdf).

9. Dunstan DW, Kingwell BA, Larsen R, et al. Breaking up prolonged sitting reduces postprandial glucose and insulin responses. *Diabetes Care* 2012;35(5):976–83.
10. Dunstan DW, Zimmet PZ, Welborn TA, et al. The Rising Prevalence of Diabetes and Impaired Glucose Tolerance: The Australian Diabetes, Obesity and Lifestyle Study. *Diabetes Care* 2002;25(5):829–34.
11. Duivivier BMFM, Schaper NC, Bremers MA, et al. Minimal Intensity Physical Activity (Standing and Walking) of Longer Duration Improves Insulin Action and Plasma Lipids More than Shorter Periods of Moderate to Vigorous Exercise (Cycling) in Sedentary Subjects When Energy Expenditure Is Comparable. *PLoS One* 2013;8(2):e55542.
12. Ebara T, Kubo T, Inoue T, et al. Effects of adjustable sit-stand VDT workstations on workers' musculoskeletal discomfort, alertness and performance. *Ind Health* 2008;46(5):497–505.
13. Ekelund U, Steene-Johannessen J, Brown WJ, et al. Does physical activity attenuate, or even eliminate, the detrimental association of sitting time with mortality? A harmonised meta-analysis of data from more than 1 million men and women [Internet]. *Lancet* 2016;388(10051):1302–10. Available from: [http://dx.doi.org/10.1016/S0140-6736\(16\)30370-1](http://dx.doi.org/10.1016/S0140-6736(16)30370-1)
14. Freedson PS, Melanson E, Sirard J. Calibration of the Computer Science and Applications, Inc. accelerometer. *Med Sci Sport Exerc* 1998;30(5):777–81.
15. Hamilton M, Hamilton D, Zderic T. Role of low energy expenditure and sitting in obesity,

- metabolic syndrome, type 2 diabetes, and cardiovascular disease. *Diabetes* 2007;56(11):2655–67.
16. Hansen RK, Andersen JB, Vinther AS, et al. Breaking up Prolonged Sitting does not Alter Postprandial Glycemia in Young , Normal-Weight Men and Women. *Int J Sport Med* 2016;
 17. Henson J, Davies MJ, Bodicoat DH, et al. Breaking Up Prolonged Sitting With Standing or Walking Attenuates the Postprandial Metabolic Response in Postmenopausal Women: A Randomized Acute Study. *Diabetes Care* 2016;39(1):130–8.
 18. Hermens HJ, Freriks B, Disselhorst-Klug C, Rau G. Development of recommendations for SEMG sensors and sensor placement procedures. *J Electromyogr Kinesiol* 2000;10(5):361–74.
 19. Júdice PB, Hamilton MT, Sardinha LB, Zderic TW, Silva AM. What is the metabolic and energy cost of sitting, standing and sit/stand transitions? *Eur J Appl Physiol* 2015;116(2):263–73.
 20. Katzmarzyk PT. Standing and mortality in a prospective cohort of canadian Adults. *Med Sci Sports Exerc* 2014;46(5):940–6.
 21. Lin Y-H, Chen C-Y, Cho M-H. Effectiveness of leg movement in reducing leg swelling and discomfort in lower extremities. *Appl Ergon* 2012;43(6):1033–7.
 22. McGill SM, Hughson RL, Parks K. Lumbar erector spinae oxygenation during prolonged contractions: implications for prolonged work. *Ergonomics* 2000;43(4):486–93.
 23. Miyashita M, Park JH, Takahashi M, Suzuki K, Stensel D, Nakamura Y. Postprandial lipaemia: Effects of sitting, standing and walking in healthy normolipidaemic humans. *Int J Sports Med* 2013;34(1):21–7.

24. Pesola AJ, Laukkanen A, Tikkanen O, Finni T. Heterogeneity of Muscle Activity during Sedentary Behavior. *Appl Physiol Nutr Metab* 2016;41(11):1155–62.
25. Roberts S, Desbrow B, Grant G, Anoopkumar-Dukie S, Leveritt M. Glycemic response to carbohydrate and the effects of exercise and protein. *Nutrition* 2013;29(6):881–5.
26. Romijn JA, Coyle EF, Sidossis LS, et al. Regulation of endogenous fat and carbohydrate metabolism in relation to exercise intensity and duration. *Am J Physiol Endocrinol Metab* 1993;265(3):e380-391.
27. Ross R, Jackson A. Exercise concepts, calculations, and computer applications. *Med Sci Sport Exerc* 1991;23(7):887.
28. Sedentary Behaviour Research Network. Letter to the editor: standardized use of the terms “sedentary” and “sedentary behaviours”. *Appl Physiol Nutr Metab* 2012;37(3):540–2.
29. Spriet LL. New insights into the interaction of carbohydrate and fat metabolism during exercise. *Sport Med* 2014;44(1):87–96.
30. Thorp AA, Kingwell BA, Sethi P, Hammond L, Owen N, Dunstan DW. Alternating bouts of sitting and standing attenuate postprandial glucose responses. *Med Sci Sports Exerc* 2014;46(11):2053–61.
31. Van, Chey T, Ding D, Chau JY, Stamatakis E, Bauman AE. Standing time and all-cause mortality in a large cohort of Australian adults. *Prev Med (Baltim)* 2014;69:187–91.

FIGURE LEGENDS

Figure 1. Timeline of the measurement day. After the preparatory phase (45 minutes quiet sitting), fasting blood samples were taken before glucose loading (0 min). Blood samples were then retaken at 30, 60, and 120 min.

Figure 2. Responses (mean \pm SD) of glucose (a), insulin (b), triglyceride (c), glycerol (d), FFA (e) and cortisol (f) to a standardized glucose loading (75g) during sitting and standing at work for 120 minutes.

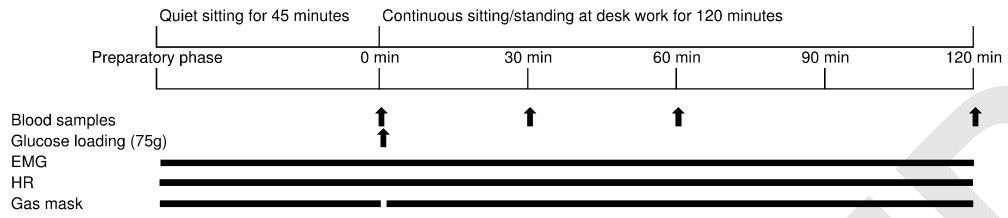
Figure 3. Fuel switching when standing up from sitting after glucose loading: fat oxidation was increased and carbohydrate (CHO) usage was reduced. This figure refers to the findings for energy expenditure (EE), muscle activity and blood samples.

LIST OF SUPPLEMENTAL DIGITAL CONTENT

Supplemental Digital Content 1.pdf—muscle activity of different muscle groups,

<http://links.lww.com/MSS/A929>

Figure 1



ACCEPTED

Figure 2

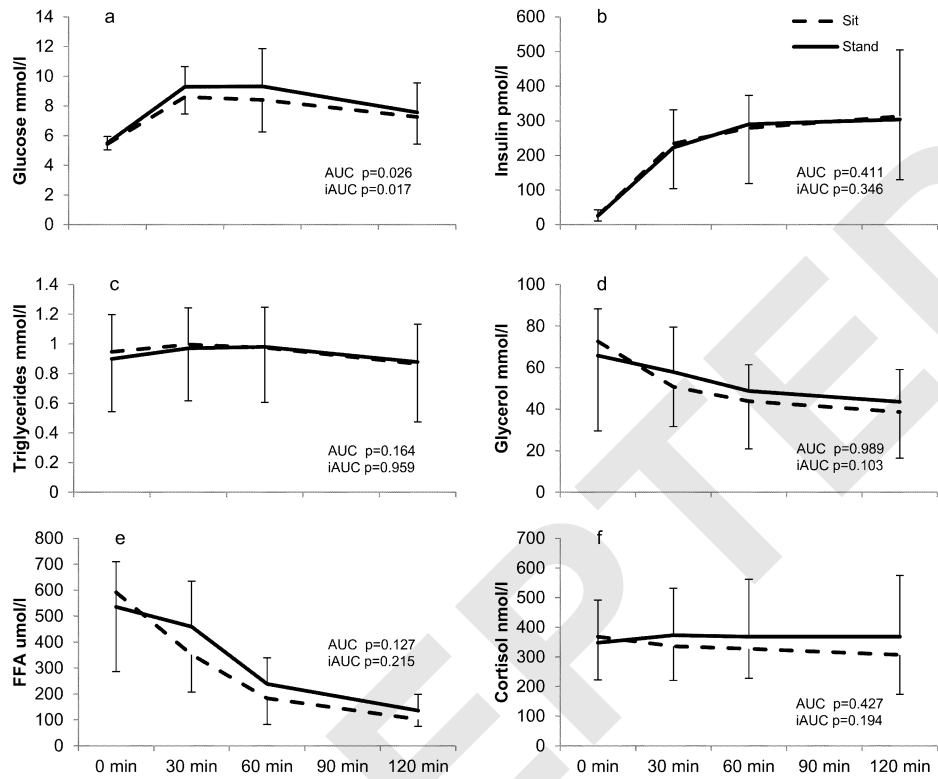


Figure 3

Fuel switching when standing up from sitting

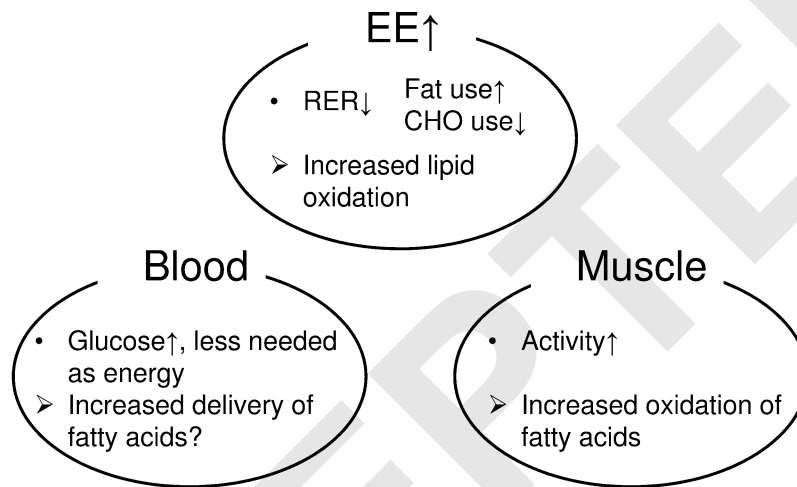


Table 1. Subject characteristics

Variables (n = 18)	Means \pm SD
Age (y)	49.4 \pm 7.9
Height (cm)	164.6 \pm 7.2
Body mass (kg)	63.2 \pm 7.8
BMI (kg/m ²)	23.4 \pm 2.8
Skeletal muscle mass (kg)	25.0 \pm 2.7
Body free fat mass (kg)	45.5 \pm 4.5
Body fat mass (kg)	17.7 \pm 6.5
Percent body fat (%)	27.5 \pm 7.1
Sitting time at work (%)	73.1 \pm 19.9
Standing time at work (%)	14.3 \pm 13.1
Walking time at work (%)	12.7 \pm 11.1
Leisure sitting time (h)	3.4 \pm 1.2
	% (n)
Education above college level	83.3 (15)
Self-rated health	
very good or rather good	88.9 (16)
average	11.1 (2)
Physical fitness level ^a	
low (0–1)	16.7 (3)
medium (2–3)	38.9 (7)
high (4–7)	44.4 (8)
Computer use at work	
more than 4 hours per day	83.3 (15)
2–4 hours	16.7 (3)
Duration of continuous computer use	
< 1 hour	11.1 (2)
1–2 hours	22.2 (4)
more than 2 hours	27.8 (5)
alternate between short and long periods	38.9 (7)
Breaks during sitting	
several times a day	61.1 (11)
less than once a day	33.3 (6)
never	5.6 (1)

^a Non-exercise physical activity questionnaire classified by category I (0–1) as low, category II (2–3) as medium, category III (4–7) as high (27).

ACCEPTED

Table 2. Baseline assessments in fasting condition (0 min) on the two experimental days

Variables (n = 18)	Sit	Stand	p values
Body mass (kg)	63.4 ± 7.7	63.4 ± 7.8	0.966
Physical activity (min/day) ^a			
recording time	884.4 ± 81.4	879.2 ± 91.7	0.605
sedentary	579.8 ± 111.7	569.2 ± 106.3	0.628
light intensity	269.1 ± 75.6	272.6 ± 108.4	0.877
moderate-to-vigorous	36.6 ± 27.8	38.2 ± 25.0	0.764
Sleep time (min/day)	480.5 ± 50.4	467.3 ± 48.5	0.338
Dietary intakes			
EE (kcal/day)	1786.6 ± 331.5	1802.6 ± 411.5	0.827
fat (g/day)	70.7 ± 23.0	68.7 ± 23.1	0.583
proteins (g/day)	80.4 ± 19.4	75.9 ± 26.8	0.318
carbohydrate (g/day)	190.3 ± 54.7	202.5 ± 50.7	0.435
Fasting condition			
Total cholesterol (mmol/l)	4.98 ± 0.92	4.89 ± 0.87	0.363
HDL-C (mmol/l)	1.95 ± 0.45	1.94 ± 0.45	0.571
LDL-C (mmol/l)	2.85 ± 0.8	2.77 ± 0.7	0.279
Triglycerides (mmol/l)	0.95 ± 0.25	0.90 ± 0.35	0.084
Glycerol (mmol/l)	72.6 ± 43.1	65.8 ± 22.5	0.519
FFA (umol/l)	591.8 ± 306.4	535.1 ± 174.8	0.528
Cortisol (nmol/l)	368.4 ± 145.9	348.0 ± 143.6	0.616
Glucose (mmol/l)	5.4 ± 0.4	5.5 ± 0.4	0.428
Insulin (pmol/l)	26.7 ± 16.0	25.7 ± 14.7	0.679
HR (bpm)	67.4 ± 10.6	65.7 ± 8.8	0.167

^a Missing n = 2

Abbreviations: EE, energy expenditure; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; FFA, free fatty acids

Table 3. Muscle activity, energy expenditure, heart rate and metabolic biomarkers during sitting and standing protocols.

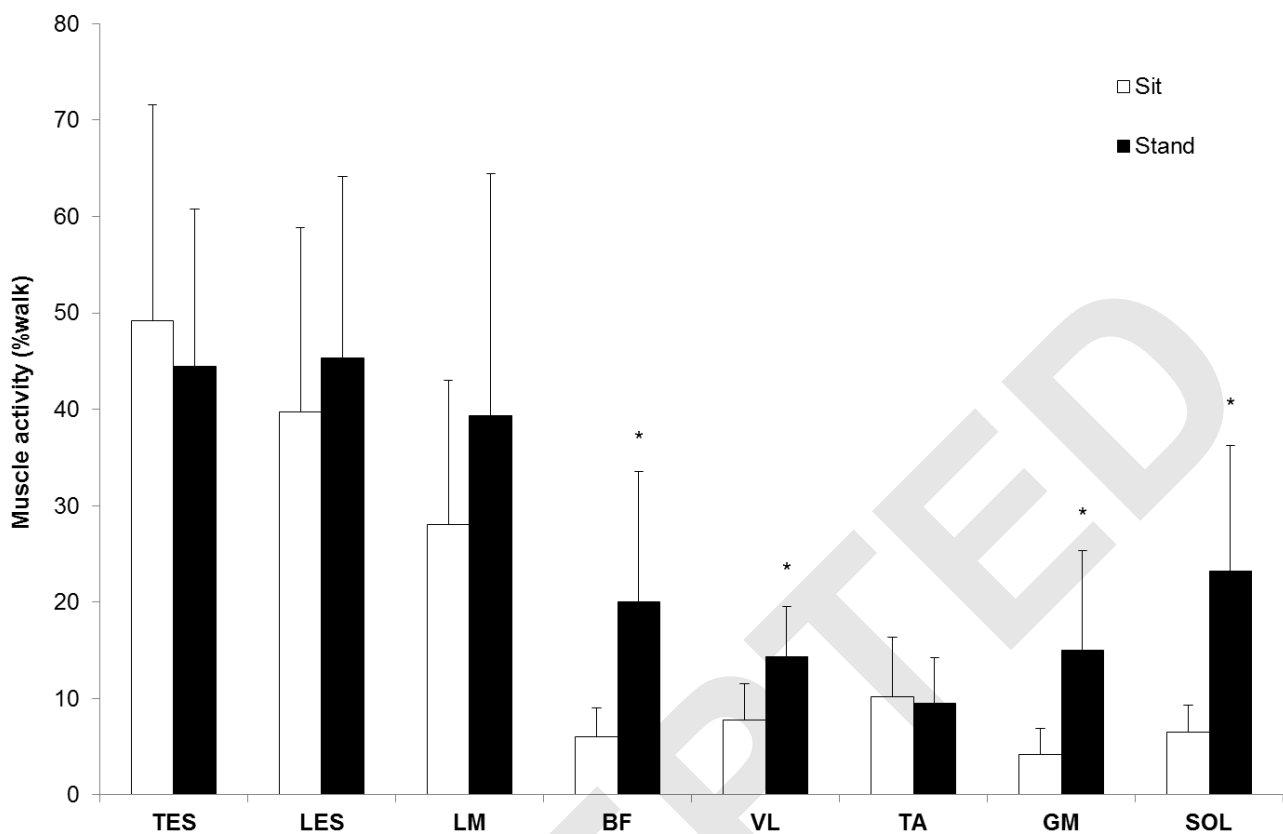
Variables (n = 18)	Sit	Stand	p values
EMG activity (% walk)			
Overall	19.1 ± 5.9	26.4 ± 9.4	0.006
Back	39.0 ± 16.6	43.0 ± 18.4	0.446
Thigh	6.9 ± 2.1	17.2 ± 8.4	0.000
Leg	7.0 ± 2.5	15.9 ± 6.1	0.000
Energy expenditure ^a			
VE (L/min)	8.5 ± 1.3	9.5 ± 1.8	0.002
BF (L/min)	15.3 ± 2.2	16.3 ± 2.7	0.037
VO ₂ (ml/min)	226.4 ± 28.9	248.3 ± 35.4	0.001
VCO ₂ (ml/min)	199.2 ± 25.5	212.1 ± 33.3	0.031
VO ₂ /kg (ml/min·kg ⁻¹)	3.6 ± 0.5	4.0 ± 0.6	0.001
RER	0.879 ± 0.021	0.853 ± 0.026	0.005
MET	1.0 ± 0.2	1.1 ± 0.2	0.001
EE (kcal/min)	1.1 ± 0.1	1.2 ± 0.2	0.002
Fat (%EE)	39.4 ± 7.3	48.3 ± 9.1	0.008
Carbohydrate (%EE)	60.6 ± 7.3	51.7 ± 9.1	0.008
HR (bpm)	75.0 ± 12.6	83.8 ± 14.8	0.000
Metabolic markers ^a			
tAUC Glucose (mmol/L·min)	897.7 ± 139.4	981.7 ± 182.5	0.026
iAUC Glucose	246.7 ± 125.0	321.7 ± 159.6	0.017
tAUC Insulin ^b (pmol/L·min)	28512.2 ± 11812.0	30135.1 ± 16423.5	0.411
iAUC Insulin ^b	25006.6 ± 10297.5	26912.1 ± 15310.9	0.346
tAUC Triglycerides (mmol/L·min)	112.5 ± 32.8	112.4 ± 46.4	0.989
iAUC Triglycerides	0.9 ± 6.0	5.7 ± 12.4	0.103
tAUC Glycerol (mmol/L·min)	5676.7 ± 2188.9	6263.2 ± 1688.4	0.164
iAUC Glycerol	-2274.6 ± 2313.0	-1765.6 ± 2214.0	0.959
tAUC FFA (umol/L·min)	29925.7 ± 11444.1	36019.5 ± 11183.4	0.127
iAUC FFA	-37932.7 ± 28876.0	-27300.8 ± 21020.1	0.215
tAUC Cortisol ^b (nmol/L·min)	38539.5 ± 11369.3	42799.3 ± 21261.4	0.427
iAUC Cortisol ^b	-5524.5 ± 11621.0	-0.7 ± 11426.9	0.194

* Bold p values indicate a significant difference between conditions.

^a Missing n = 2

^b Missing n = 3

Abbreviation: VE, ventilation; BF, breath frequency; tAUC, total area under curve; iAUC, net incremental area under curve



Supplemental Figure 1. Muscle activity of thoracic erector spinae (TES), lumbar erector spinae (LES), lumbar multifidus (LM), biceps femoris (BF), vastus lateralis (VL), tibialis anterior (TA), gastrocnemius medialis (GM) and soleus (SOL) during sitting and standing at work. * Significant difference based on paired t-tests ($p < 0.01$).