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# Electromyography of Sedentary Behavior: Identifying Potential for Cardiometabolic Risk Reduction

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<sup>1</sup>Active Life Lab, South-Eastern Finland University of Applied Sciences, Mikkeli, FINLAND; <sup>2</sup>Baker Heart & Diabetes Institute, Melbourne, VIC, AUSTRALIA; <sup>3</sup>The University of Queensland, School of Human Movement and Nutrition Sciences, Brisbane, QLD, AUSTRALIA; <sup>4</sup>Deakin University, Institute for Physical Activity and Nutrition (IPAN), School of Exercise and Nutrition Sciences, Melbourne, VIC, AUSTRALIA; <sup>5</sup>Centre for Urban Transitions, Swinburne University of Technology, Melbourne, VIC, AUSTRALIA; and <sup>6</sup>Faculty of Sport and Health Sciences, Neuromuscular Research Center, University of Jyväskylä, FINLAND

## ABSTRACT

LAMBERG, S., C. J. BRAKENRIDGE, D. W. DUNSTAN, T. FINNI, G. N. HEALY, N. OWEN, and A. J. PESOLA. Electromyography of Sedentary Behavior: Identifying Potential for Cardiometabolic Risk Reduction. *Med. Sci. Sports Exerc.*, Vol. 57, No. 1, pp. 11–22, 2025. **Introduction:** Muscle activation during interruptions to prolonged sedentary time is a hypothesized mechanism underlying observed cardiometabolic benefits. We examined associations of quadriceps and hamstring muscle activity patterns with cardiometabolic risk markers and how these patterns varied between different sitting-interruption countermeasures. **Methods:** Electromyographic (EMG) data (shorts) were gathered for 1 to 2 d from healthy adults in a free-living study ( $n = 172$ , age  $40.9 \pm 12.9$ , BMI  $23.6 \pm 1.3$ ) and a laboratory-based study ( $n = 12$ , age  $47.0 \pm 7.7$ , BMI  $30.0 \pm 4.7$ ). Patterns examined were average EMG (aEMG;%EMG<sub>MVC</sub>); EMG activity duration (% above signal baseline 3  $\mu$ V); and usual (weighted medians) EMG activity bout amplitude (%EMG<sub>MVC</sub>) and duration (s). In the free-living study, these were regressed against risk markers (waist, fat percentage, fasting plasma glucose, total cholesterol, high-density lipid cholesterol, low-density lipid cholesterol, triglycerides); in the laboratory study, EMG patterns for the muscle groups were compared between sitting and the active countermeasures. **Results:** In the free-living study, lower-extremity muscles displayed minimal overall activity, with hamstrings and quadriceps using only 2.6% and 2.0% of their capacity (%EMG<sub>MVC</sub>), respectively, and being active for 30% and 25% of the time. Higher hamstring aEMG and EMG activity duration were beneficially associated with waist, high-density lipid cholesterol and fat percentage (duration only) and a longer quadriceps usual EMG activity bout duration was beneficially associated with fasting plasma glucose. In the laboratory study, compared with prolonged sitting, active seated or upright active-interruption countermeasures modified these EMG patterns; brief (6 min) walking and simple resistance activities (SRA) were more beneficial than was a bout of standing (30 min) with the SRAs being the only intervention that matched daily aEMG levels. **Conclusions:** Upright and physically active interruptions to sitting appear to be required to increase the typically low muscle engagement observed in free-living contexts, promoting muscle activity patterns that may help ameliorate cardiometabolic risk. **Key Words:** ELECTROMYOGRAPHY, MUSCLE CONTRACTILE ACTIVITY, CARDIOMETABOLIC HEALTH

**T**ime spent sitting (sedentary behavior) can be associated adversely with aspects of cardiometabolic health risk (1). Sedentary behavior is defined as any waking

behavior characterized by an energy expenditure <1.5 times the basal metabolic rate, that is, 1.5 metabolic equivalent of task (MET), while in a sitting, lying or reclining posture (2). Laboratory-based trials have evaluated sedentary behavior countermeasures and their effects on cardiometabolic risk markers in the acute experimental setting. Examples of these include replacing sitting time with body weight resistance exercises (3–5), walking (6,7), stair climbing (8), fidgeting (9), and pedaling while sitting down (7). Benefits of regularly interrupting sedentary time have been observed on multiple metabolic health indicators, including insulin (3,10,11), and glucose control (3,11,12). Effective and pragmatic countermeasures, such as those that would be applicable to common high-volume sitting domains like typical office work (12,13), are needed to better understand how to address the health risks associated with prolonged periods of sedentary behavior (14). Although there is evidence of these countermeasures being individually effective for cardiometabolic risk markers, there have been less studies directly comparing them, and comparing

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them according to their proposed mechanism of benefit: muscle contraction.

Increased muscle activity of the lower extremities (e.g., gluteal muscle group, hamstrings, quadriceps) is associated with improved glycemic control (15,16). Compared with the muscle inactivity and altered homeostasis that can characterize sitting time, the metabolic demand when muscles are contracted can increase by up to 100-fold (17). This increases both contraction-mediated glucose uptake, and blood flow at the vicinity of muscle fibers, which are key drivers of enhanced whole body glucose tolerance (18). Different types of muscle activity and different muscle groups can have distinct effects on these mechanisms. For example, experimental studies have shown that stationary cycling increases insulin sensitivity mainly in the quadriceps muscles, compared with hamstrings (19). In participants with type 2 diabetes or prediabetes, a greater rectus femoris glucose uptake was observed during sprint interval running as compared with moderate-intensity running (20). Sustained activity in the soleus muscle, which is characterized by high proportion of oxidative type 1 fibers, increased both local oxidative metabolism and whole-body glucose regulation in the laboratory setting (21). Reducing sedentary behavior by at least 0.5 h, was associated with increased insulin sensitivity in hamstring muscles but not quadriceps, which was speculated to be a result of increased activity when transiting to and holding upright postures (22). Collectively, these findings suggest that there may be site, intensity, and activity type variations in the biologically beneficial impacts of muscle activity patterns. Therefore, quantifying muscle specific activation patterns during sitting countermeasures and in daily living can inform on mechanisms and assist in designing sedentary behavior interventions.

The degree to which muscle contractile capacity is used is often characterized by electromyography (EMG) amplitude, which can be reported relative to muscle maximal voluntary contraction, or MVC, over an entire recording period (23,24). This total EMG amplitude is occurring through EMG bouts, which can be further characterized in terms of their continuous duration and amplitude patterns. For example, an activity EMG bout can be accumulated in a long duration but with low overall amplitude, or short duration and high overall amplitude. Especially relevant to sedentary behavior interventions is the duration of contractile time occurring (25). Distinguishing these pattern metrics within sedentary behavior countermeasures may offer new mechanistic perspectives, with (for example) potential implications for contraction-mediated glucose uptake (18). Although it is generally understood that reducing and interrupting prolonged sitting is beneficial, the pattern of muscular contraction (duration and amplitude) within the breaking up of muscular inactivity, and the associations of these patterns with cardiometabolic risk markers, are unknown.

We aimed to address these evidence gaps using data collected from both free-living and laboratory-based protocols. The first aim was to explore associations of free-living quadriceps and hamstring muscle activity patterns with cardiometabolic risk biomarkers to identify potentially-fruitful muscle-activation intervention targets. The second aim was to compare muscle

activity patterns between different prolonged sitting-interruption countermeasures, to gain further insight as to their potential efficacy in sedentary behavior intervention.

## METHODS

Data were drawn from three studies: two free-living studies (pooled data from the EMG24 (26) and InPact (27) studies, collectively referred to as “habitual study”) and an experimental laboratory study (OPTIMUS pilot, referred to as “laboratory study”). The EMG24 project was approved by the Ethics Committee of the University of Jyväskylä, the InPact project was approved by the Ethics Committee of the Central Hospital District of Central Finland, and the Laboratory study was approved by the Human Research Ethics Committee of the Hospital District of Northern Savo (475/13.02.00/2021). All participants provided written informed consent.

### Habitual Study Design and Protocol

The aim of the habitual study was to quantify free-living quadriceps and hamstring muscle activity patterns and examine associations of EMG activity patterns with cardiometabolic markers in a free-living setting. We pooled cross-sectional data from two separate studies; the EMG24 study conducted in 2007 to 2012 (26) and the InPact study (27) conducted in 2011 and in 2013. EMG24 (26) was a cross-sectional study to quantify muscle loading during normal daily life, whereas the InPact study (27) was a sedentary time-targeted randomized controlled trial. Only baseline data were used from the InPact study.

Methods for these two studies have been previously published (15,26,28). In brief, participants without history of chronic disease, not pregnant, that had one full day of EMG measurement (at least 8 h of wear) were selected, resulting in 226 (EMG24,  $n = 109$ ; InPact,  $n = 117$ ) participants. In both studies, participants fasted for a minimum of 10 h before presenting in the morning to the laboratory for body composition and biochemical assessment. Pathology-measured fasting plasma glucose (FPG), high-density (HDL) and low-density (LDL) lipid cholesterol, and triglycerides using standardized procedures (Konelab 20 XT analyzer; ThermoFisher, Espoo, Finland). Following this, participants performed a battery of isometric exercises to normalize EMG signals to MVC. Participants were then instructed to go about their daily lives, wearing the EMG shorts underneath their normal clothing.

### Laboratory Study Design and Protocol

The aim of the laboratory study was to quantify and compare quadriceps, hamstring, and gluteal muscle activity patterns between different prolonged sitting interruption countermeasures. The selected countermeasures have been found to effectively improve cardiometabolic health outcomes, but potentially differ in their muscle activity patterns and may provide alternative pragmatic and feasible ways to interrupt prolonged sitting in the office environment (4,6,7,29–31). The study included a short (~1 h) introduction visit and a 1-d laboratory experiment

(~8 h), in which participants were asked to test different conditions while working at an office desk (Fig. 1).

**Laboratory study recruitment and screening.** Participants were recruited during June and October of 2021. Recruitment occurred through nearby health clinics and via social media. Before participation, participants completed an electronic eligibility survey. Participants were eligible if they were age between 35 and 65 yr, had a body mass index (BMI) between 25 and 50 kg·m<sup>2</sup>, worked at least 0.8 full-time equivalent in a desk-based occupation, and worked in the Mikkeli or Savonlinna area. Exclusion criteria were as follows: pregnancy; currently using a height adjustable workstation at their workplace (and reported standing more than 50% of working time); regularly engaged in moderate-intensity exercise  $\geq 30$  min·d<sup>-1</sup> for >3 months; regularly engaged in >30 min·wk<sup>-1</sup> of structured strength/resistance training (i.e., involving machine or free weights) for >3 months; regularly sitting for <6 h·d<sup>-1</sup> for >3 months; major illness/physical problems (acute or chronic) that may limit participation in the intervention; unable to communicate in Finnish; and, unable to provide written informed consent. This cohort represents those more likely to benefit from sitting-reduction interventions. Final eligibility was confirmed with each participant by phone call with research personnel.

A sample size of 12 was required to detect a significant difference between the sitting condition and the countermeasures at a two-sided 0.05 significance level with an 82% probability, if the true difference between conditions is 1.5%EMG<sub>MVC</sub>. This assumes that the within-subject standard deviation in EMG amplitude is 1.1%EMG<sub>MVC</sub> (28).

Figure 1 shows the study visit schedule and experimental condition protocols. Fifteen volunteers completed the eligibility survey. Two participants were excluded either based on standing too much at work or having a BMI <25. One participant withdrew from the study, resulting in a final study sample of 12.

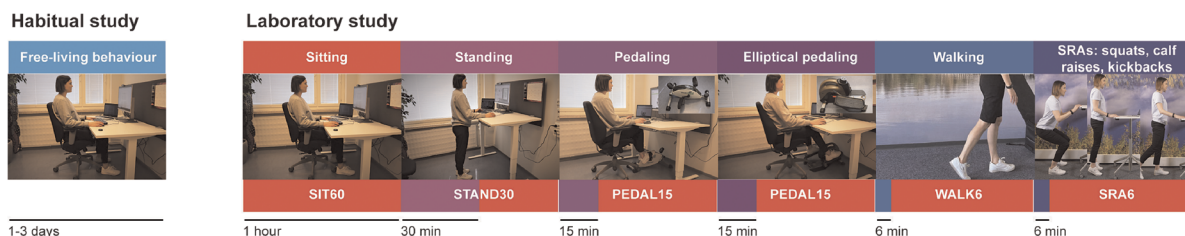
On the experimental day, participants had their weight, height, and BMI assessed with standard procedures. Participants were then instructed to wear a pair of tight-fitting EMG shorts (sizes XL and XXL, Myontec Ltd., Kuopio, Finland). Sitting, standing, pedaling, elliptical pedaling, walking and simple resistance activities were first measured for 3 min for warm-up and familiarization and to quantify EMG activity. Next, isometric maximal voluntary contractions (MVC) for each muscle group were measured to normalize the EMG signal (%EMG<sub>MVC</sub>) according to

Myontec Ltd MVC field testing recommendations. Maximal voluntary contractions for left and right quadriceps were measured in seated isometric knee extension. Maximal voluntary contractions for left and right hamstrings and gluteal muscles were measured in upright isometric hip extension. Two warm-up contractions (verbally instructed to be 50% effort level from the maximum) were performed for each movement (left and right). These were followed by two 5-s maximal contractions, with a 1-min break between the MVCs. In the analysis phase, the MVC with the higher EMG amplitude (1-s average) was used for signal normalization.

**Experimental sitting interruption conditions.** Experimental conditions were performed on 1 d in a randomized order over 6 h. Before participants started each condition, the researcher gave verbal and showed written instructions on how to perform them. The duration of sitting to activity ratio was selected to standardize the MET level between active conditions based on prior research (7,32). Total workload for all conditions was controlled at approximately 2.5 MET hours (by modifying the ratio of sitting to activity duration) to investigate how EMG activation patterns differ between different countermeasures breaking up prolonged sitting at the same overall energy expenditure (33). In addition to the MET-level, the duration of conditions was selected based on demonstrated efficacy in improving glycemic control. Previous studies have reported that interrupting prolonged sitting every 40 to 60 min with 3 to 15 min of simple resistance activities, walking or pedaling have effectively improved glycemic control (4,7). Moreover, interrupting sitting every 54 min with a 6-min bout of simple resistance activities was more effective in improving glucose 7-h iAUCnet than interrupting sitting every 27 min with a 3-min bout (4). We employed a single bout of 6 min of walking and simple resistance activities. Experimental conditions were:

**SIT60:** Participants sat for 1 h and were instructed to work like they usually do while sitting. This condition was considered as the reference condition. MET value for occupational sitting is defined to be 1.5 (33).

**STAND30:** Participants first stood for 30 min and then sat 30 min at a height-adjustable sit-stand desk. They were instructed to stand as they normally would while engaging in a work task. While standing, the researcher ensured that the participant did not take any steps.



**FIGURE 1—Habitual and laboratory study designs.** In the laboratory study, the activity of quadriceps, hamstring, and gluteal muscle groups was measured with EMG-sensing shorts during a seated desk-work control condition (sitting 60 min [SIT60]), and during five interruption conditions within a 60-min period: standing 30 min (STAND30); active sitting with pedals 15 min (PEDAL15); active sitting with elliptical pedals 15 min (ELL15); walking 6 min (WALK6); and simple resistance activities 6 min (SRA6). The activity of quadriceps and hamstring muscle groups was measured in free-living settings in the habitual study.

**PEDAL15:** Participants completed 15 min of under-desk pedaling (DeskCycle2; 3D Innovations LLC, Greeley, CO) followed by sitting with feet on the ground for 45 min. Participants were instructed to pedal at 50 to 60 rounds per minute (supervised by researcher), and with a corresponding workload of 30 to 40 W.

**ELL15:** Participants completed 15 min of elliptical pedaling (Exerpeutic 900E, NetFit Europe) and sat 45 min. Participants were instructed to pedal at 50 to 60 revolutions per minute corresponding to 30 to 40 W.

**WALK6:** Participants completed 6 min of walking with the remaining 54 min seated. Participants walked in unobstructed clinic hallways and were encouraged to walk at a comfortable, purposeful pace.

**SRA6:** Participants completed 6 min of simple resistance activities (squats, standing calf raises and leg kickbacks) and sat 54 min. They were instructed to perform each activity for 20 s and then move onto the next activity, repeating this process six times.

**Randomization.** Participants were randomly assigned to complete the six sitting interruption conditions via a balanced block randomization. Randomization was performed before the first day of measurement. The randomization sequence was generated with an online tool ([www.randomization.com](http://www.randomization.com)).

### Habitual and Laboratory EMG Measurement and Sensor Data Synthesis

For all studies, knitted fabric shorts were used to measure EMG from the skin surface of the quadriceps, hamstring muscles and then also gluteal muscles in laboratory study (Myontec Ltd., Kuopio, Finland). The reference electrodes of the shorts were placed longitudinally on the lateral sides of the left and right thighs on the covering membranes of the iliotibial tract (28), the bipolar measuring electrodes were situated on the distal region of the quadriceps and hamstring muscles and on the middle of the gluteal muscles.

The EMG signal was stored in a 50-g electronic module attached to the waist. The signal was recorded with a sampling frequency of 1000 Hz, band-pass filtered at 40 to 200 Hz (−3 dB), digitalized with a 24-bit A/D converter and a gain of 0, averaged with nonoverlapping windows of 40 ms (to 25 Hz) and saved in the module. The data was downloaded and visualized with the Muscle Monitor software (Myontec Ltd, Kuopio, Finland) and lab log timestamps were revised based on the visualized signal. Conditions were identified from continuous timeseries with recorded timestamps. Next, the individual channels from the right and left quadriceps, hamstring and gluteal muscles were normalized to the respective  $EMG_{MVC}$  (% $EMG_{MVC}$ ). The contraction with the higher amplitude was used for normalization (1 s mean of the peak activation phase). The signal was further smoothed with a 200-ms moving average algorithm (25,34).

EMG baseline can sometimes drift (26). Possible drift was corrected by subtracting the minimum value of a moving

5-min window from each data point preceding the window (25,26,28). The normalized, smoothed, baseline filtered data were plotted and visually checked for artifacts. In the instance of a prolonged artifact, the channel was removed. The EMG shorts have demonstrated validity, repeatability, responsiveness, and feasibility, and detailed descriptions of the recording devices have been reported previously (24–26).

### EMG Amplitude and Pattern of Accumulation

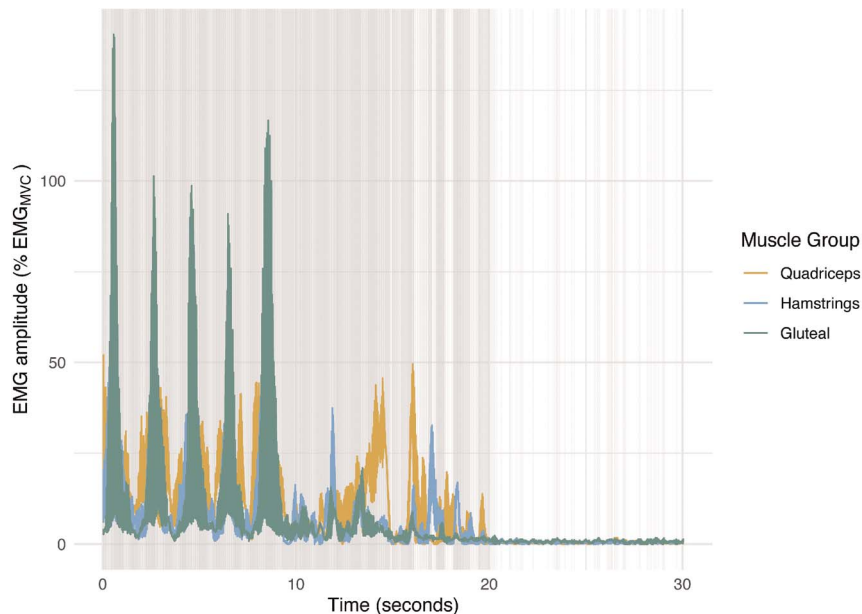
Electromyography activity variables were calculated across the entire waking period for the habitual study, and sequentially for each condition in the laboratory study. In the instance that participants had more than one valid day ( $n = 94$ ) of habitual EMG observation, the EMG activity variables were averaged across the observation days.

Electromyography amplitude was analyzed as percentage of EMG during maximal voluntary isometric contraction (% $EMG_{MVC}$ ) over the entire recording period (Fig. 2). Electromyography activity accumulates from EMG bouts, which can have different duration and amplitude (Fig. 2). An EMG activity bout is defined as a period when EMG amplitude is continuously above the signal baseline (3  $\mu$ V, Fig. 2). The activity threshold 3  $\mu$ V was selected because it provides the best responsiveness and is not confounded by body composition (25). The duration of an EMG activity bout represents the time that thigh and gluteal muscle groups work continuously without a rest period, and the amplitude of an EMG bout is the mean amplitude of this continuous bout. Quantifying the pattern of how EMG accumulates is important because both duration and intensity of activation can regulate the glucose metabolism pathways at the muscle fiber level (18).

The EMG signal consists of a very high number of very short and low intensity activity bouts, causing a positively skewed density. Therefore, some typical summary metrics, such as the mean or median, do not represent the data well. As suggested by Chastin and Granat (35), we used a nonlinear regression technique (Levenberg-Marquart) to fit a sigmoid function  $\frac{t^n}{(t^n + dW_{50\%}^n)}$ , where  $t$  is the EMG bout duration,  $n$  a free parameter, and  $dW_{50\%}$  the usual EMG activity bout duration, above or below which 50% of EMG duration is accumulated. A similar approach was used to calculate usual EMG activity bout amplitude, above and below which 50% of EMG amplitude is accumulated. However, because EMG bout duration affects the contribution of each bout to EMG amplitude, the duration was used as a weight in the calculations.

### Statistical Analyses

Analyses were conducted using R (R version 4.2.2; R Foundation for Statistical Computing, Vienna, Austria). In the habitual study, muscle activity variables were regressed with cardiometabolic risk biomarkers. Linear regression models were made for a combined model where quadriceps and hamstrings were in the same model (primary analysis model) and for hamstrings and quadriceps separately. All regression models were adjusted for daily minutes of EMG wearing time, age, and sex.



**FIGURE 2**—Muscle group-specific normalized EMG-signal sample during transition from SRAs to sitting. An EMG bout was defined for each muscle group as a continuous period when the signal was above the EMG bout threshold (gray areas). Usual EMG activity bout amplitude and duration were quantified for each EMG bout (weighted median bout amplitude and duration only within the gray area), and EMG amplitude was calculated over the entire recording period (gray and white areas) to quantify overall use of capacity.

Further adjustment for fat percentage was made for combined model. Regression results were reported as unstandardized coefficients ( $\beta$ ). A variance inflation factor (VIF) was tested to check for multicollinearity.

In the laboratory study analyses, experimental conditions were compared with generalized linear mixed effects models using random slopes and intercepts. The active conditions were compared with the reference condition SIT60 to inform pattern changes when actively interrupting sitting, and to STAND30 to inform pattern changes between stationary vs. dynamic interruptions. Log transformation of dependent variables improved normality of residuals. When comparing activity conditions, we first proposed two models: one with and one without muscle group interaction and compared model fits using ANOVA. Muscle group interaction had lower BIC and was selected. Results are presented as relative rates (RR), with the reference condition value set to 1. Accordingly, a relative rate of 0.7 would signify that the estimated value is 30% lower compared with the reference value. For all analyses statistical significance was set at  $P < 0.05$  (two-tailed).

## RESULTS

On average, the habitual study sample was younger ( $40.9 \pm 12.9$ ) and had a lower BMI ( $23.6 \pm 1.3$ ) compared with laboratory study sample ( $47.0 \pm 7.7$  yr;  $30.0 \pm 4.7$  kg·m<sup>2</sup>). The habitual study participants were healthy, with no known diseases and with biomarkers on average within normal range (FPG  $5.3 \pm 0.5$ , total cholesterol  $4.9 \pm 0.9$ , HDL cholesterol  $1.8 \pm 0.5$ , LDL cholesterol  $2.6 \pm 0.8$  and triglycerides  $1.0 \pm 0.8$ ). The habitual study EMG wear time was on average  $1.8 \pm 1.7$  d,  $12.9 \pm 1.3$  h·d<sup>-1</sup>.

Multiple linear regression results for the associations of hamstring and quadriceps habitual EMG patterns with

cardiometabolic risk markers are shown in Table 1, and separately in Supplemental Table 1 (Supplemental Digital Content, <http://links.lww.com/MSS/D79>) for univariate quadriceps, hamstrings, and average channel relationships. Multiple linear regression results further adjusted for fat percentage are shown in Supplemental Table 2 (Supplemental Digital Content, <http://links.lww.com/MSS/D79>). Estimated marginal means of quadriceps, hamstring and gluteal muscles EMG patterns in habitual and laboratory studies are visualized in Figure 3 and the corresponding numerical values are reported in Supplemental Table 3 (Supplemental Digital Content, <http://links.lww.com/MSS/D79>). Differences between quadriceps and hamstring EMG patterns within the laboratory conditions are shown in Supplemental Tables 4 and 5 (Supplemental Digital Content, <http://links.lww.com/MSS/D79>) by using relative rates. In the following paragraphs, the cardiometabolic risk markers associations, habitual, and laboratory patterns are summarized by the EMG pattern variable.

### aEMG

During habitual living, the hamstring aEMG (%EMG<sub>MVC</sub> [95%CI]: 2.6 [2.3, 2.9]) was higher as compared with the quadriceps aEMG (%EMG<sub>MVC</sub> [95%CI]: 2.0 [1.7, 2.2],  $P < 0.001$ , Figure 3). Considering the combined model, a higher hamstring aEMG than quadriceps aEMG was associated with lower waist circumference. Similarly, a higher hamstring aEMG than quadriceps aEMG was associated with higher HDL cholesterol (Table 1). After further adjustment for fat percentage, a higher quadriceps aEMG than hamstring aEMG was associated with lower fasting plasma glucose, albeit with a smaller effect size (Supplemental Table 2, Supplemental Digital Content, <http://links.lww.com/MSS/D79>).

TABLE 1. Linear regression results between habitual EMG patterns and cardiometabolic risk biomarkers for combined quadriceps and hamstring relationships ( $n = 172$ ).

	Quadriceps			Hamstrings		
	$\beta$	95% CI	<i>P</i>	$\beta$	95% CI	<i>P</i>
<b>aEMG (%EMG<sub>MVC</sub>)</b>						
Waist	1.21	-0.11 to 2.53)	0.07	-1.55	-2.68 to -0.43)	<b>0.01</b>
Fat%	0.70	-0.28 to 1.68	0.16	-0.74	-1.58 to 0.11	0.09
FPG	-0.05	-0.11 to 0.01	0.12	-0.03	-0.08, 0.02	0.28
Total cholesterol	-0.01	-0.13 to 0.11	0.88	0.03	-0.07 to 0.14	0.52
HDL	-0.01	-0.04 to 0.02	0.49	0.03	0.00-0.06	<b>0.03</b>
LDL	0.01	-0.03 to 0.06	0.52	-0.01	-0.05 to 0.02	0.43
Triglycerides	-0.01	-0.06 to 0.04	0.71	-0.02	-0.07 to 0.02	0.31
<b>EMG activity duration (%)</b>						
Waist	0.23	0.01-0.45	<b>0.04</b>	-0.31	-0.51 to -0.10	<b>0.01</b>
Fat%	0.03	-0.13 to 0.19	0.75	-0.21	-0.37 to -0.06	<b>0.01</b>
FPG	-0.01	-0.02 to 0.00	0.27	-0.01	-0.01 to 0.00	0.28
Total cholesterol	0.00	-0.02 to 0.02	0.99	0.01	-0.01 to 0.03	0.53
HDL	0.00	-0.01 to 0.00	0.35	0.01	0.00 to 0.01	<b>0.03</b>
LDL	0.00	-0.01 to 0.01	0.75	0.00	-0.01 to 0.01	0.96
Triglycerides	0.00	-0.01 to 0.01	0.90	-0.01	-0.02 to 0.00	0.06
<b>Usual EMG activity bout amplitude (%EMG<sub>MVC</sub>)</b>						
Waist	0.04	-0.41 to 0.49	0.86	-0.24	-0.66 to 0.18	0.26
Fat%	0.14	-0.18 to 0.47	0.39	-0.05	0.26-0.76	0.76
FPG	-0.01	-0.03 to 0.01	0.18	-0.01	-0.03 to 0.01	0.37
Total cholesterol	0.02	-0.02 to 0.06	0.42	0.01	-0.03 to 0.05	0.67
HDL	0.00	-0.01 to 0.01	0.56	0.01	0.00-0.02	0.22
LDL	0.01	0.00-0.02	0.20	-0.01	-0.02 to 0.01	0.41
Triglycerides	0.00	-0.00 to 0.01	0.60	0.00	-0.02 to 0.01	0.78
<b>Usual EMG activity bout duration (s)</b>						
Waist	0.01	-0.20 to 0.31	0.66	-0.25	-0.52 to 0.02	0.07
Fat%	0.00	-0.01 to 0.01	0.81	-0.01	-0.02 to 0.01	0.41
FPG	-0.01	-0.20 to 0.00	<b>0.05</b>	0.00	0.00 to 0.01	0.84
Total cholesterol	-0.01	-0.30 to 0.02	0.54	0.00	-0.01 to 0.1	0.73
HDL	0.00	-0.01 to 0.00	0.88	0.00	0.00-0.01	0.06
LDL	0.00	0.00-0.01	0.76	0.00	0.00-0.00	0.64
Triglycerides	0.00	-0.01 to 0.00	0.34	0.00	-0.01 to 0.00	0.59

\* Note: Regression results are presented as unstandardized coefficients ( $\beta$ ) with 95% CI.

In the laboratory study, as compared with SIT60, all sitting interruption conditions increased aEMG, but there were no differences in quadriceps and hamstring aEMG increments. However, PEDAL15 (RR [95% CI]: 0.7 [0.5, 0.9]) and ELL15 (RR [95% CI]: 0.6 [0.4, 0.9]) had an estimated 30–40% lower gluteal activity as compared with quadriceps activity (Supplemental Tables 3 and 4, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 3).

Compared with STAND30, only WALK6 and SRA6 increased aEMG. However, WALK6 (RR [95% CI]: 0.8 [0.7, 1.0]) and SRA6 (RR [95% CI]: 0.7 [0.6, 0.9]) increased hamstring aEMG 20–30% less, and PEDAL15 (RR [95% CI]: 0.6 [0.5, 0.8]), ELL15 (RR [95%CI]: 0.6 [0.5, 0.8]), WALK6 (RR [95%CI]: 0.8 [0.6, 1.0]), and SRA6 (RR [95%CI]: 0.7 [0.6, 1.0]) increased gluteal aEMG 20–40% less, as compared to quadriceps changes. (Supplemental Tables 3 and 5, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 3)

### EMG Activity Duration

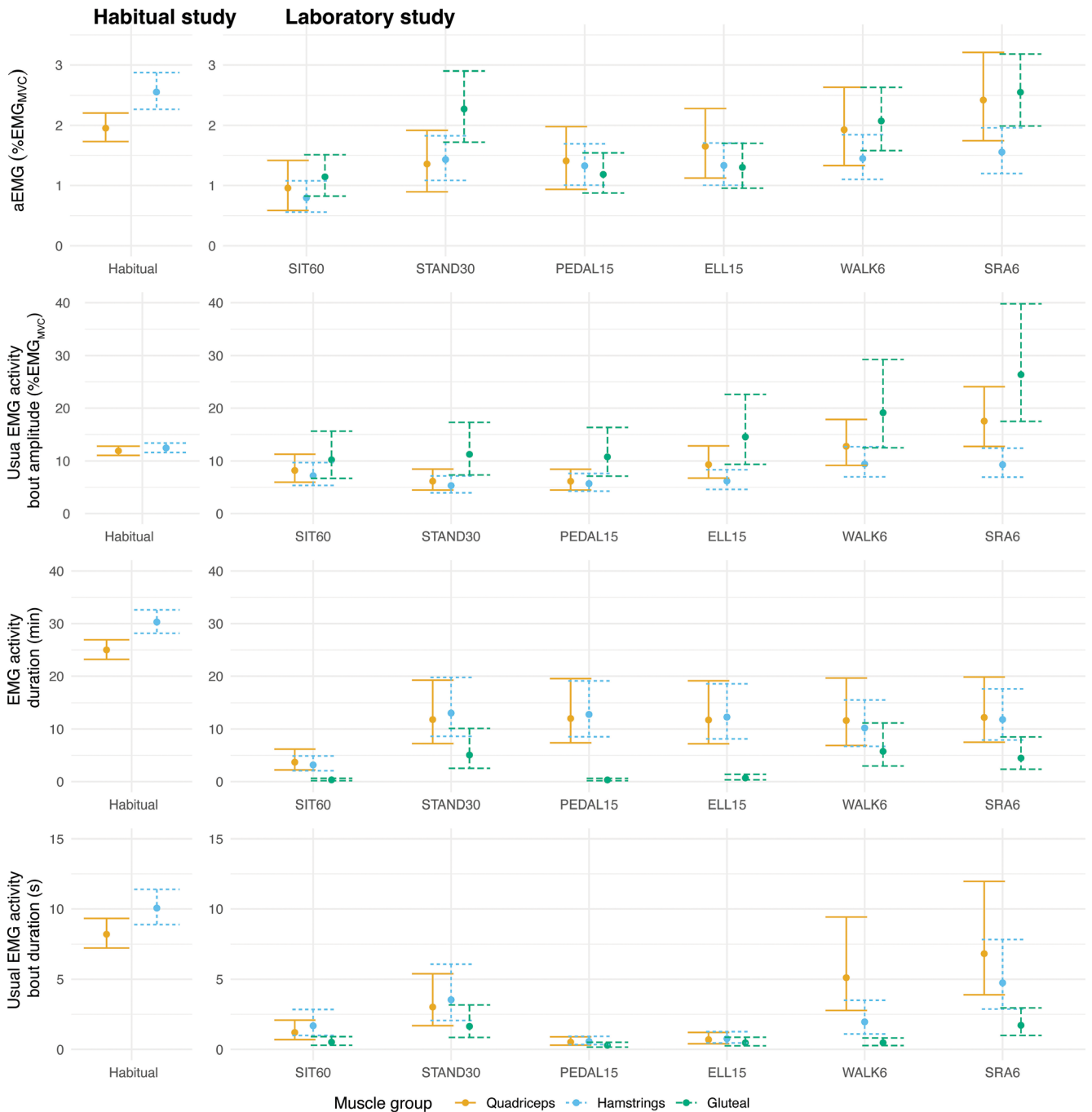
The habitual living hamstring EMG activity duration was longer (% [95% CI]: 30.3 [28.2, 32.6]) as compared to the quadriceps duration (% [95% CI]: 25.0 [23.2, 26.9]  $P < 0.001$ , Figure 3). A longer hamstring EMG activity duration, than quadriceps EMG activity duration, was associated with a lower waist circumference, fat percentage and HDL (Table 1). After further adjustment for fat percentage, these associations were

no longer statistically significant (Supplemental Table 2, Supplemental Digital Content, <http://links.lww.com/MSS/D79>).

All sitting interruption conditions increased EMG activity duration compared with SIT60, with no differences between quadriceps and hamstring muscle groups. However, STAND30 (RR [95% CI]: 4.9 [2.1, 11.5]), WALK6 (RR [95%CI]: 5.7 [2.4, 13.6]) and SRA6 (RR [95% CI]: 4.2 [1.8, 9.6]) increased gluteal EMG activity duration 420% to 570% more, and PEDAL15 (RR [95% CI]: 0.3 [0.1, 0.7]) increased gluteal EMG activity duration 70% less, as compared with quadriceps changes (Supplemental Tables 3 and 4, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 2). Compared with STAND30, PEDAL15 (RR [95% CI]: 0.1 [0.03, 0.1]) and ELL15 (RR [95% CI]: 0.1 [0.1, 0.3]), resulted in a 90% lower gluteal EMG activity duration than quadriceps (Supplemental Tables 3 and 5, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 3).

### Usual EMG Activity Bout Amplitude

The habitual living hamstring usual EMG activity bout amplitude (%EMG<sub>MVC</sub> [95% CI]: 12.5 [11.6, 13.4]) was higher as compared to the quadriceps muscle group (%EMG<sub>MVC</sub> [95% CI]: 11.6 [11.1, 12.8]  $P < 0.001$ , Figure 3). There were no associations between the muscle group specific usual EMG activity bout amplitude and any of the cardiometabolic risk markers (Table 1).



**FIGURE 3—Differences between quadriceps, hamstring and gluteal muscles EMG patterns in habitual and laboratory studies visualized as estimated marginal means.**

All sitting interruption conditions except ELL15 increased the usual EMG activity bout amplitude as compared with SIT60. The SRA6 (RR [95% CI]: 0.6 [0.4, 0.9]) increased hamstring usual EMG activity bout amplitude 40% less than that of quadriceps increment (Supplemental Tables 3 and 4, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 3). Compared with STAND30, ELL15, WALK6, and SRA6 increased usual EMG activity bout amplitude. However, SRA6 (RR [95%CI]: 0.6 [0.4, 0.9]), increased hamstring usual EMG activity bout amplitude 40% less as compared

with quadriceps (Supplemental Tables 3 and 5, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 3).

### Usual EMG Activity Bout Duration

The habitual living hamstring usual EMG activity bout duration was longer (s [95% CI]: 10.06 [8.87, 11.40]) as compared with the quadriceps duration (% [95% CI]: 8.21 [7.23, 9.32]  $P < 0.001$ , Figure 3). A longer quadriceps usual EMG activity bout duration, as compared to the hamstrings, was



associated with a lower FPG (Table 1). After further adjustment for fat percentage, a longer hamstring usual EMG activity bout duration, than quadriceps, was associated with a higher HDL (Supplemental Table 2, Supplemental Digital Content, <http://links.lww.com/MSS/D79>).

All sitting interruption conditions except ELL15 increased the usual EMG activity bout duration. Compared to SIT60, WALK6 increased hamstring (RR [95%CI]: 0.3 [0.1, 0.7]), and gluteal (RR [95%CI]: 0.2 [0.1, 0.6]) usual EMG activity bout duration 70% to 80% less as compared with quadriceps (Supplemental Tables 3 and 4, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 3). Compared with STAND30, PEDAL15 and ELL15 decreased, and SRA6 increased usual EMG activity bout duration. Compared with STAND30, WALK6 increased hamstring (RR [95% CI]: 0.3 [0.1, 0.8]) and gluteal (RR [95% CI]: 0.2 [0.1, 0.5]) usual EMG bout duration 70% to 80% less as compared with quadriceps increment (Supplemental Tables 3 and 5, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 3).

## DISCUSSION

Muscle contractile activity is one of the key postulated mechanisms of physical activity benefit (1,15,16). The present study directly quantified quadriceps and hamstring contractile patterns, investigated their associations with cardiometabolic risk markers, and compared the efficacy of different sitting interruption countermeasures on modifying these patterns. In summary, only less than 3% of muscles contractile capacity was used during daily living, with the muscle groups examined being active for 30% of measurement time. Average EMG and EMG activity duration in hamstrings, compared with quadriceps, was beneficially associated with waist circumference, fat percentage and HDL cholesterol. In contrast, a higher usual EMG activity bout duration in quadriceps, compared with hamstrings, was beneficially associated with FPG. All sitting interruption conditions, regardless of posture, beneficially modified the EMG activity patterns. However, upright postures with ambulation (walking and simple resistance activities) were most efficacious in increasing hamstring aEMG, and quadriceps usual EMG activity bout duration, and were the only ones matching the low habitual aEMG level. This suggests specificity in associations of different quadriceps and hamstring muscles EMG patterns with cardiometabolic risk markers and identifies how these may be modified with different sitting interrupting countermeasures.

The findings show that hamstring and quadriceps activity patterns can be differently associated with cardiometabolic risk markers. These different associations can potentially be explained by the functional and physiological differences between these muscle groups. The hamstrings are crucial for stabilizing the hip and knee joints and essential for maintaining an upright posture during everyday activities. The quadriceps, being the primary extensors of the knee, are more involved in activities that require dynamic movement, such as walking or climbing stairs (36,37). A previous laboratory study found that

reducing sedentary behavior improved insulin sensitivity in the hamstring muscles, but not in the quadriceps, of inactive and overweight individuals (22). This effect was thought to result from increased activity levels associated with walking and maintaining upright postures at light to moderate intensities (22). In the present study, the hamstring muscle group was more active than the quadriceps during daily living (both in terms of aEMG, EMG activity duration and usual EMG activity bout amplitude), which might be the main reason for hamstring activity more effectively contributing to the beneficial biomarker profile (including waist, HDL and fat percentage). However, further adjustment for fat percentage attenuated the associations with hamstring EMG activity duration (but not those with hamstring aEMG). In contrast, only a longer quadriceps usual EMG activity bout duration, and after adjustment for fat percentage also quadriceps aEMG, was beneficially associated with a lower FPG concentration.

In the laboratory study, only the dynamic upright countermeasures (WALK6 and SRA6) increased usual EMG activity bout duration and amplitude more in the quadriceps than hamstring muscle groups. This might indicate that in the healthy habitual study cohort, higher intensity physical activities, requiring increased quadriceps activation, were required for a beneficial association with FPG concentration. After further adjustment to fat percentage, a higher quadriceps aEMG and usual EMG activity bout duration, as compared with hamstrings, were also beneficially associated with a lower FPG concentration, which again emphasizes the role of quadriceps activity for this outcome. Nevertheless, because of the exploratory nature of the additional analyses and suggestive properties of cross-sectional findings, we cannot rule out reverse-causality and confounding in the findings. For example, greater hamstring activity can be indicative of a greater overall activity, not necessarily reflecting specific or unique hamstring-related benefits. Further controlled experimental research is needed to confirm our inferences.

Another distinction between the quadriceps and hamstring muscle groups are the known differences in fiber type profiles. Both hamstring and quadriceps muscle fiber type compositions are described as oxidative, glycolytic, or mixed, but the contribution of these different fiber types during physical activity may differ (38,39). In hamstrings, there might be a potential pool of fibers, capable of transformation either to slow type 1 or to fast type 2a to tune the functional response especially in biceps femoris (39). Notably, hamstrings exhibit higher resistance to fatigue compared to quadriceps, suggesting the predominant utilization of, or adaptation, to using the oxidative fibers in typical-use cases, making it more resistant to fatigue, and well-suited for activities involving sustained contractions (40). The quadriceps muscles fiber type composition is mixed but predominantly consists of phasic fibers that are activated during more intense physical activity (19,20). Therefore, although the hamstrings and quadriceps share similar compositional features, they may demonstrate differing metabolic tendencies during daily activities. (41).

Meta-analyses of acute laboratory studies have demonstrated that physically active interruptions to sitting can mitigate

postprandial increases in glucose and insulin levels (29,42). However, these studies have not confirmed the distinct impact of activity intensity or type on these effects. The finding that breaking up prolonged sitting with upright and physically active interruptions were most efficacious in changing the EMG patterns that were beneficially linked with cardiometabolic risk markers is supported by emerging experimental evidence. Simple resistance activities (3 min every 30 min or 6 min every 60 min) have been found to reduce postprandial blood glucose and insulin responses compared with sitting (3–5). Light-intensity walking (2–5 min every 20–60 min) has demonstrated similar positive effects in overweight or obese participants (7,43). In our findings, SRAs and walking were more effective in increasing aEMG as compared with standing or pedaling, despite a longer duration and a similar estimated overall mean MET-level between the conditions. Given that sedentary behavior interventions outside of the laboratory setting have resulted only in modest average improvements in body anthropometry, glucose metabolism, lipid metabolism, and blood pressure (1,44), our findings support the emerging view that more active countermeasures to prolonged sitting (beyond brief interruptions and standing) should be prioritized to increase daily muscle activity and to reach meaningful cardio-metabolic benefits (44).

In our study, interrupting 60 min of sitting with 30 min of standing significantly increased average EMG amplitude, EMG activity duration and usual EMG bout duration relative to sitting. However, the increments in aEMG were smaller, and in EMG activity duration similar (the EMG outcomes most consistently associated with cardiometabolic risk markers), to the dynamic alternatives walking and SRAs. Previous laboratory studies have found that standing can modestly improve postprandial glucose and insulin responses in persons with overweight or obesity, but not necessarily in those of normal weight (6,29,30,45). Moreover, previous EMG studies have shown that standing EMG amplitude is higher in overweight than normal weight individuals suggesting that muscles of those who are overweight must do more work to hold up the greater weight (46). One implication arising from the present findings is that a longer period of standing may be required to increase EMG amplitude to a similar degree to the more intense countermeasures. Because the MET-level was matched between the conditions, the longer standing period would mean that a higher energy expenditure is required from a standing countermeasure to reach a similar aEMG and potentially metabolic benefits, as compared with walking or SRAs. However, extending the duration of standing bouts may not enhance vascular and metabolic markers to the same extent as brief, yet more frequent interruptions (47). These findings collectively suggest that achieving a cardiometabolically beneficial EMG profile through standing may be more challenging compared with more dynamic activities, such as walking and SRAs.

A strength of our study is the direct measurement of EMG activity with EMG shorts, which provide even better repeatability compared with traditional bipolar electrodes since they capture EMG from a larger area (24). A novel element is the examination of the patterning of EMG activity under different conditions (laboratory setting; habitual living). This is important

since EMG activity bout characteristics (duration and volume of amplitude) can influence contraction-related energetic pathways, and this study suggests these EMG-derived outcomes are linked with cardiometabolic risk markers.

However, although the use of EMG is the explicit strength of this study, several factors need to be considered when comparing EMG activity between individuals. It should be noted that EMG measurements of the present study represent only the superficial muscle groups located below the electrodes (48) and are not representative of other lower- or upper-extremity muscles that are also important for upright and ambulatory movement. Other lower leg muscles, particularly those related to weight-bearing and ambulation (like the soleus), can improve insulin sensitivity and glucose regulation (21). Methodological studies have shown that an EMG inactivity threshold above signal baseline (3  $\mu$ V), as used in this study, provides overall the best responsiveness indices (25). Further, correlations between EMG inactivity duration and leg fat mass, and fat percentage, were weak (25). However, we cannot rule out that body composition, and particularly fat tissue thickness below the recording electrodes, may influence EMG amplitude and be a potential confounder of the analyses (49), and needs to be considered when interpreting the findings. A potential limitation is that electrode shorts do not measure EMG of just one muscle at a time affecting the usual EMG activity duration outcome. It may be the case that usual EMG activity bout duration has variability because the thigh electrodes are sampling across several muscles with somewhat different functions, especially since the rectus femoris is a two-joint muscle. These “burst analysis” outcomes have been reported in previous EMG work with the aim to report EMG burst (or bout, as we have identified) characteristics, from which the total EMG duration and amplitude eventually accumulates from (26). However, many of the burst outcomes are highly skewed and hence comparing their mean metrics may not be statistically sound (25). Previous accelerometer studies show that the accumulation pattern of activity may be important beyond the total activity (50). For example, a person can accumulate 8 h of sitting from two 4-h bouts, or from 16  $\times$  30-min bouts, with potentially different cardiometabolic effects. Such EMG accumulation outcomes may provide important insights and open new avenues that can extend upon the findings reported from accelerometer studies. The high variance seen during walking may be one such finding: some people may relax their muscles during walking, whereas others may have a more tense walking pattern, which ultimately results in a high variance in this outcome.

The measurement duration was relatively short due to the EMG shorts data logging capacity. However, previous investigations have reported adequate test–retest repeatability for similar EMG measurement durations (25,28), high intra-individual variability in sitting (as high as 4.5 h) has been reported over 7 d of accelerometry use (51) and it is unknown how “typical” the day chosen of EMG wear was for the participant. In future, extending the wear period (as possible) will likely improve the reliability and estimate of daily EMG levels. The MVC testing

protocol was different between the habitual and laboratory study and may inhibit the comparison of absolute aEMG and usual EMG activity bout amplitude comparison between the studies. However, we expect this did not prevent comparing the relative quadriceps and hamstring activation patterns. There was some artifact in the data, which is typical for EMG measurements. However, removal of individual channels has a limited influence on the extracted EMG outcomes (25).

Active countermeasure durations in laboratory setting were assigned an intensity level based on the rate of energy expenditure (EE) expressed as METs (33). MET values for pedaling and SRA were obtained from previous studies that defined them in the context of prolonged sedentary behavior (7). However, even though durations of countermeasures were assigned based on EE, we did not explicitly measure the activities with calorimetry methods and therefore exact matching of energy expenditure was not achieved. Although this increases external validity, the actual energy expenditure between the laboratory study active countermeasures is not necessarily the same.

There were some differences in gluteal muscle group activation in the laboratory study, but the EMG shorts in the habitual study were not equipped with gluteal electrodes. The laboratory study indicates that gluteal muscles might play a significant role in response to different sitting interruption conditions. The activation of the gluteal muscles during sitting interruption conditions were relatively strong in relation to their maximum but occurred in shorter bouts (Supplemental Table 4, Supplemental Digital Content, <http://links.lww.com/MSS/D79>; Fig. 3). The variations in EMG activity duration, and amplitude of gluteal muscles suggest their sensitivity to different physical activities and interruptions, highlighting their importance in musculoskeletal dynamics and possible associations for cardiometabolic risk markers which should be investigated in the future. During seated pedaling muscle contractile activity was low, especially in the gluteal muscles. The result may be affected by the fact that the electrodes in the area of the gluteal muscles and partially in the hamstring muscles were attached to an external surface, i.e., the chair.

## CONCLUSIONS

In conclusion, these findings suggest specificity in how different quadriceps and hamstring muscle EMG patterns are associated with cardiometabolic risk markers and elucidate how these patterns can be modified with different sitting-interruption

countermeasures. The daily hamstring and quadriceps muscle engagement was very low, below 3% of  $EMG_{MVC}$ . Specific muscle activity patterns, including elevated hamstring average EMG and prolonged hamstring EMG activity duration, were beneficially associated with waist circumference, fat percentage, and HDL levels. Conversely, extended usual EMG activity bout duration in the quadriceps was beneficially associated with fasting plasma glucose levels. Although the respective sitting interruption regimens were designed to have the same energy expenditure, only the upright and physically active interruptions elevated the aEMG level to match the low daily muscle engagement. These findings offer important insights for designing targeted interventions to interrupt sitting and strategies to mitigate the health impacts of sedentary behavior, indicating that upright and physically active interruptions are the most effective muscular countermeasures against sedentary time.

## AUTHOR CONTRIBUTIONS

S.L. participated in the conception and design of experiments, data collection, assisting of data analysis, drafting of article, approved final version of the article. C,J,B. participated in the conception and design of experiments, data analysis, drafting of the article, approved final version of the article. D.W.D. participated in the conception and design of experiments, drafting of article, revision of article, approved final version of the article. T.F. participated in the conception and design of experiments, revision of article, approved final version of the article. G.N.H. participated in the conception and design of experiments, drafting of article, approved final version of the article. N.O. participated in the conception and design of experiments, drafting of article, revision of article, approved final version of the article. A.J.P. participated in the conception and design of experiments, secured funding, data analysis, drafting of article, revision of article, approved final version of the article.

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