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Title of the manuscript:

Objectively measured absolute and relative physical activity intensity levels in postmenopausal women

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Abstract

Objectives. To investigate how objectively measured physical activity (PA) levels differ according to absolute moderate intensity recommendation (3-6 METs) and relative to individual lactate thresholds (LT1 and LT2), and to verify if high-fit women record higher PA levels compared to women with lower aerobic fitness. **Methods.** Seventy-five postmenopausal women performed an incremental exercise test and several constant-velocity tests wearing an accelerometer to identify the activity counts ($\text{ct}\cdot\text{min}^{-1}$) corresponding to LT1 and LT2. Individual linear regression determined activity counts cut-points for each intensity: 1) sedentary ($<200 \text{ ct}\cdot\text{min}^{-1}$), 2) light (from $200 \text{ ct}\cdot\text{min}^{-1}$ to $\text{ct}\cdot\text{min}^{-1}$ at LT1), 3) moderate ($\text{ct}\cdot\text{min}^{-1}$ between LT1 and LT2) and 4) vigorous ($\text{ct}\cdot\text{min}^{-1} >\text{LT2}$). Participants then wore an accelerometer during a week to measure the time spent at each PA intensity level. **Results.** According to absolute intensity categorization, high-fit postmenopausal women recorded twice as much time at moderate-to-vigorous PA (MVPA) ($P < 0.01$) than low-fit women. However, when PA intensity was calculated relative to individual lactate thresholds, MVPA was significantly reduced by half ($P < 0.01$) and the data revealed no differences ($P = 0.62$) between groups ($\sim 20 \text{ min}\cdot\text{day}^{-1}$ at MVPA). **Conclusions.** Accelerometer cut-points derived from absolute moderate-intensity recommendations overestimated MVPA. Similar time at MVPA was recorded by high- and low-fit postmenopausal women when the cut-points were tailored to

individual lactate thresholds. A more accurate estimation of PA behavior could be provided with the use of individually tailored accelerometer cut-points.

Keywords: Accelerometry; cardiorespiratory fitness; individually tailored cut-points; moderate-to-vigorous physical activity.

Introduction

Physical activity (PA) level and cardiorespiratory fitness (CRF) are two of the most powerful predictive factors of premature death and chronic disease (Arem et al., 2015; Kodama et al., 2009). Observational prospective cohort studies using self-reported questionnaires suggest that meeting current PA guidelines (American College of Sports Medicine, 2013; Garber et al., 2011; Physical Activity Guidelines Advisory Committee, 2018) of 150 min·wk⁻¹ of moderate-to-vigorous physical activity (MVPA) reduces all-cause mortality risk by 26-31% (Arem et al., 2015; Lee, Rexrode, Cook, Manson, & Buring, 2001; Wen et al., 2011). Several studies have reported that MVPA, rather than total energy expenditure or light intensity PA is determinant to reduce the levels of cardiometabolic risk factors (Arem et al., 2015; Swain & Franklin, 2006; Weston, Wisloff, & Coombes, 2014) and osteoporosis (Whitfield, Kohrt, Pettee Gabriel, Rahbar, & Kohl, 2015).

Large epidemiology studies using self-reported PA questionnaires (Arem et al., 2015; Wen et al., 2011) have commonly grouped recreational PA into three intensity categories according to their energy requirements, supported by evidence-informed guideline recommendations (Ainsworth et al., 2011; American College of Sports Medicine, 2013; Garber et al., 2011; Pate et al., 1995; Physical Activity Guidelines Advisory Committee, 2018): light (<3 metabolic equivalents [MET]), moderate (3-6 METs) and vigorous (>6 METs). Interestingly, Lee and colleagues (Lee, Sesso, Oguma, & Paffenbarger Jr, 2003) showed that it is the relative intensity of exercise, rather than total energy expended or absolute intensity of activities what causes greater reduction in coronary heart disease (CHD) mortality risk. During the last years, accelerometers have replaced PA questionnaires to objectively capture human activity, applying activity cut-points derived from the traditional absolute 3-6 MET intensities to obtain representative volumes of MVPA (Freedson, Melanson, & Sirard, 1998; Santos-Lozano et al., 2013; Sasaki, John, & Freedson, 2011). These studies have generated widely

divergent regression models for converting activity counts to energy expenditure, yielding different cut-points for PA intensity categories (Migueles et al., 2017). Contrary to self-reported estimates, results of accelerometer-based objective assessments of PA report much lower volumes of MVPA (Arem et al., 2015; Tucker, Welk, & Beyler, 2011) indicating that only a small proportion of adults (7-15%) meet current PA recommendations (Cerin et al., 2014; Colley et al., 2011; Kujala et al., 2017; Tucker et al., 2011). Reported objective weekly volumes of MVPA are significantly lower in women and vary from as low as seven $\text{min}\cdot\text{day}^{-1}$ to 57 $\text{min}\cdot\text{day}^{-1}$ (Cerin et al., 2014; Colley et al., 2011; Kujala et al., 2017; Tucker et al., 2011). Previous studies have demonstrated that the use of fixed cut-points may under or overestimate MVPA due to the lack of consideration of an individual's CRF (Miller, Strath, Swartz, & Cashin, 2010; Ozemek, Cochran, Kaminsky, Strath, & Byun, 2013; Rejeski et al., 2015). When moderate intensity activity counts cut-points have been calculated based on relative intensities (*i.e.*, 45-60% $\dot{V}O_{2\text{max}}$) the results show substantial differences in measured activity counts between less fit and most fit individuals (Miller et al., 2010; Ozemek et al., 2013). Furthermore, Kujala and colleagues (2017) using heart-rate based PA assessment reported that although the time spent at MVPA applying fixed absolute intensity cut-points was higher in men compared to women, and decreased with age, when intensity levels were calculated relative to individual's CRF, these differences disappeared.

The validity of this "relative intensity" approach has also been substantially criticized because exercising at the same relative intensity could elicit a wide range of metabolic stress across individuals (Meyer, Gabriel, & Kindermann, 1999). The determination of individual physiological break points of energy supply, as the lactate thresholds (LT), is the gold standard method for accurate exercise intensity prescription (Beneke, 2003; Binder et al., 2008; Weltman et al., 1990). Besides, the achieved workload at LT is an accurate indicator of CRF (Faude, Kindermann, & Meyer, 2009). In this study, we were interested in accurately quantifying PA

volumes in postmenopausal women because they usually report lower MVPA compared to men (Kujala et al., 2017; Tucker et al., 2011), and because they have an increased risk of osteoporosis and cardio-metabolic diseases related to menopause, lower fitness and inactivity (Kanis et al., 2012; Kannel, Hjortland, McNamara, & Gordon, 1976). It is unknown how well individually-tailored accelerometer cut-points derived from LT reflect PA levels during routine activities of daily living, and whether differences exist between less fit and most fit women.

This study aimed to investigate how objectively measured PA levels differ according to absolute moderate intensity recommendation (3-6 METs) and relative to individual lactate thresholds (LT1 and LT2), and to verify if high-fit women record higher PA levels, especially MVPA, compared to women with lower fitness level.

Materials and Methods

Study design

This cross-sectional study investigated the volumes of absolutely and relatively (*i.e.*, relative to participant's LT) determined PA at different intensity levels (sedentary, light, moderate, vigorous, and moderate-to-vigorous combined) during one week in postmenopausal women. Participant's aerobic fitness and PA data were collected from November 2015 to June 2017. Participants performed on different days an incremental submaximal shuttle test (IST) and several constant-velocity tests (CVT) wearing an accelerometer to identify the activity counts ($\text{ct}\cdot\text{min}^{-1}$) corresponding to LT1 and LT2 velocities. Participants then wore an accelerometer for seven complete days to assess their PA patterns.

Study participants

Participants were recruited through advertisements placed on healthcare centers. One hundred and four participants were screened by telephone, 88 were deemed eligible and were

invited to participate in the study. Inclusion criteria were: 1) surgical or natural menopause (no menstrual periods during previous 12 months), 2) age <75 years. Participants were excluded from the study if they had any of the following conditions that might interfere with exercise testing: 1) presence of spine or low-trauma fractures or severe arthrosis at the hip, knees or feet, 2) functional limitation to walk for 20 minutes, 3) presence of any chronic disease that would impair the cardiorespiratory system during testing. The local hospital's ethical committee approved the study (Pyto2011/71) and written informed consent was obtained from all participants before any study procedures were undertaken. The procedure of the study was in accordance with the Declaration of Helsinki and was registered in *ClinicalTrials.gov PRS* (NCT02984553).

Exercise tests

Incremental shuttle test (IST)

Prior to the first visit, participants were instructed to abstain from caffeine and stimulants for at least four hours and strenuous activity for ≥ 24 h before testing. Height was measured using a wall stadiometer (Seca, Germany) and body mass was measured using a scale to the nearest 0.1kg (Seca, Germany). Before starting the test each participant's resting heart rate (HR) (Polar V800, Polar Electro Oy, Kempele, Finland) and blood lactate concentration ($[La^-]$) (Lactate Pro2, Arkray, Japan) were measured on a standing position. Capillary blood samples (0.3 μ L) were taken from a hyperemic earlobe. Testing was performed in a laboratory setting in a controlled temperature environment ($\sim 20^\circ$) over a 20 m indoor track. The distance of the course was extended to 20 m from the original test (Singh, Morgan, Scott, Walters, & Hardman, 1992) to keep the pace constant avoiding excessive turns that might increase the energy cost and musculoskeletal demand, potentially leading to premature fatigue, discomfort or even injury. Five cones were positioned at 0.5–5–10–15 and 19.5 m and participants had to walk in a straight line until the last cone, then turn around and return to the start (Figure 1a).

The speed was dictated by an audio signal. A double beep indicated the start of each stage. After that, participants were instructed to be at the next cone with each beep while keeping the pace as constant as possible. The IST started at 2.4 km·h⁻¹ (~2.1 METs). The intensity was progressively increased by 0.61 km·h⁻¹ (Singh et al., 1992) at each 2-min stage with 1-min rest in between. At the end of each stage, HR and [La⁻] were recorded. Each participant wore a triaxial accelerometer (Actigraph wGT3X-BT Pensacola, FL, USA) over the right iliac crest in the mid-axillary line throughout the test.

Each participant was free to start running from the 7th stage onwards (6.1 km·h⁻¹), or the operator suggested to do so when the participant was not able to match the required speed. The test was stopped when: 1) [La⁻] values were ≥ 3.0 mmol·l⁻¹ to avoid excessive fatigue, and/or 2) participant repeatedly failed to match the pace programmed, and/or 3) participant was exhausted. LT1 was defined as the highest velocity above which [La⁻] increased by an amount of ≥ 0.1 mmol·l⁻¹ in the following stage and ≥ 0.2 mmol·l⁻¹ in the subsequent stage (Figure 1b).

Constant-velocity tests (CVT)

Participants completed two to seven 20 min CVT on the same 20 m track used for the IST. Each participant performed the corresponding tests on separate testing days (one week in between). Each CVT consisted of two stages of 10 min at a constant pace with a two minutes interruption for blood sampling. Heart rate was continuously recorded, and capillary blood samples were obtained before at rest, at the 10th min and the end of exercise (22nd min). Walking or running velocity of the first CVT was programmed as the velocity at which blood lactate increased by 1 mmol·l⁻¹ above the blood lactate value at LT1 during the IST. In the following tests, the velocity was increased or decreased by ~0.30 km·h⁻¹ until the maximal lactate steady state velocity (*i.e.*, LT2) could be determined (Billat, Dalmy, Antonini, & Chassain, 1994). An increase in [La⁻] ≤ 0.4 mmol·l⁻¹ during the final 10 min of exercise was defined as steady state (Beneke, 2003) (Figure 1c).

*** Figure 1

Physical activity assessment

Each participant was instructed to wear an Actigraph wGT3X-BT accelerometer over the right iliac crest at the mid-axillary line on an elasticized belt for eight consecutive days (24h) alongside a daily log.

Each monitor was previously initialized at a 50Hz frequency and downloaded after the 8-day period in vector magnitude (VM₃) activity counts in 1-min epochs (ct·min⁻¹). Data were analyzed with Actilife 6[®] full software (Actigraph, Pensacola, FL, USA). The first recording day was not used for the analysis. Sleep periods were selected from participant's daily logs. For the present analysis, only data over the day were used. Participants were included in the analysis if they had a minimum of five days of monitoring, including two weekend days and daily wear time was ≥ 12 hours (Migueles et al., 2017). Periods of continuous zeros lasting more than 60 min with allowance for 2 min interruptions of activity counts between 0 and 200 (Troiano et al., 2008) were checked in participant's daily logs and assigned as non-wear time if corresponded (Migueles et al., 2017). High activity levels (>10.000 ct·min⁻¹) or high step counts (>20.000 steps·day⁻¹) were verified against participant's daily logs.

Accelerometer activity counts measured during the IST at the velocities corresponding to each participant's LT1 and LT2 were used to determine three individual intensity zones: light ($<LT1$ ct·min⁻¹), moderate ($LT1$ ct·min⁻¹ - $LT2$ ct·min⁻¹) and vigorous ($>LT2$ ct·min⁻¹). Both, the light-intensity and the moderate-intensity zones were subdivided into other identical two zones (low and high) (Figure 2). The time (min·day⁻¹) spent in each of these intensity zones for every valid day and the number of daily steps were averaged. Both, sedentary time and MVPA were reported in one (+1) and ten (+10) minute bouts (the minimum time required to be between the specified intensity cut-points). The time spent at each relative intensity category was

compared to absolute accelerometer activity counts cut-points, which have been previously validated against direct measurement of oxygen uptake during treadmill walking and running activities designated as movements that represent moderate intensity recommendations (3–6 METs) (*i.e.*, light = 200–2689 ct·min⁻¹, moderate = 2690–6166 ct·min⁻¹, vigorous ≥6167 ct·min⁻¹ (Sasaki et al., 2011).

Statistical analysis

Accelerometer activity counts during IST were calculated by averaging the VM₃ activity counts in one-second epochs during the central 90sec of each stage. These activity counts were then averaged in 1-min epochs (ct·min⁻¹). The corresponding ct·min⁻¹ at LT2 were selected from the individual regression equations obtained in the IST. For women who walked at the LT2 during the CVT, the data of running stages were removed. For women who ran at LT2 during the CVT and for whom this velocity was between the last walking stage and the first running stage of the IST, linear interpolation was performed.

For the whole sample, repeated measures ANOVA was used to determine differences in both, [La⁻] and activity counts throughout the IST. Bonferroni Post Hoc analysis was applied when significant effects were observed for velocity stages.

Participants were categorized into two groups according to their LT2 velocity (Low-fitness group; as ≤6.8 km·h⁻¹ [n = 37], and high-fitness group; as >6.8 km·h⁻¹ [n = 38]), which represented the median speed of LT2 and corresponded to the minimum level of CRF (6-7 METs) associated with lower event rates in 40 to 60 years old women (Kodama et al., 2009). The differences in the activity count cut-points at LT1 and LT2 between groups were presented using Tukey Box Plots and Mann Whitney U-test was used for comparison between groups.

The primary outcome of the study was the volume of objectively measured PA (particularly MVPA₊₁₀) expressed as mean ± SD, using both, relative and absolute cut-points.

Intra-group PA levels ($\text{min} \cdot \text{day}^{-1}$) were compared using a paired *t*-test or Wilcoxon test (for non-parametric data), and Independent samples test or Mann Whitney U test were used for inter-group analysis.

We conducted a post hoc power analysis using the G*Power software (Faul, Erdfelder, Lang, & Buchner, 2007). The level of statistical power reached in this study was 0.99 for the following variables; alpha level ($\alpha = 0.05$), sample size ($n = 75$), and effect size ($ES = 0.80$ relative vs. absolute MVPA₊₁₀). Statistical significance was set at $P < 0.05$. Statistical analyses were performed using SPSS statistical software (version 22.0, IBM SPSS Statistics, Chicago, IL).

Results

Population selection and characteristics

Among 104 interested participants who were screened for eligibility, 14 were excluded. Reasons for exclusion were; 1) not meeting eligibility criteria ($n = 12$), and 2) declined to participate ($n = 2$). Among 88 participants who were invited to participate, 75 completed all the assessments and were included in the study for data analysis. Reasons for exclusion in data analysis were inability, musculoskeletal pain or discomfort when running ($n = 6$), failure to get the LT2 with accuracy ($n = 3$), invalid 7-day accelerometry recording ($n = 1$), cardiovascular or pulmonary disease ($n = 2$), and failure to keep testing appointment ($n = 1$). All participants had a minimum of 5-valid days (2 weekend days included), and 78% had 7-valid days.

Determination of PA intensity levels through lactate thresholds

The average velocity vs. $[\text{La}^-]$ curve during the IST in the whole group of participants is presented in Figure 2. During the first five stages $[\text{La}^-]$ did not change noticeably. From the fifth stage onwards, $[\text{La}^-]$ significantly increased ($F_{8,568} = 137.5$, $P < 0.001$) in each subsequent

exercise stage. Accelerometer activity counts increased linearly and significantly ($F_{7,497} = 892$, $P < 0.001$) over the duration of the IST. Average LT1 was $5.1 \pm 0.7 \text{ km}\cdot\text{h}^{-1}$, and the corresponding activity counts, $[\text{La}^-]$, and HR values were $4133 \pm 1152 \text{ ct}\cdot\text{min}^{-1}$, $0.8 \pm 0.2 \text{ mmol}\cdot\text{l}^{-1}$ and $98 \pm 12 \text{ beats}\cdot\text{min}^{-1}$ (bpm), respectively. Mean LT2 was $7.1 \pm 1.0 \text{ km}\cdot\text{h}^{-1}$, and the corresponding activity counts, $[\text{La}^-]$, and HR values in the IST were $6783 \pm 2077 \text{ ct}\cdot\text{min}^{-1}$, $2.4 \pm 0.7 \text{ mmol}\cdot\text{l}^{-1}$, and $141 \pm 17 \text{ bpm}$, respectively.

*** Figure 2

Individually tailored activity counts cut-points by fitness group

High-fit women were five years younger ($P < 0.01$) and their body mass was eight kg lower ($P < 0.01$) compared to the low-fit group. LT1 and LT2 were 17% and 24% higher in high-fit compared to low-fit women ($P < 0.01$), respectively. Accordingly, measured activity counts were greater in the high fit group ($P < 0.01$) at both, LT1 (4659 ± 1250 vs. $3592 \pm 729 \text{ ct}\cdot\text{min}^{-1}$) and LT2 (7989 ± 2035 vs. $5543 \pm 1223 \text{ ct}\cdot\text{min}^{-1}$) (Figure 3).

*** Figure 3

Physical activity levels

Table 1 shows the results of the minutes spent per day at each intensity level for each group, using both, absolute intensity cut-points (Sasaki et al., 2011) and individually-tailored LT cut-points. The two groups exceeded the target of 10,000 daily steps recommended by PA guidelines (Garber et al., 2011). Recorded MVPA₊₁₀ using the absolute intensity cut-points ($31.1 \pm 24.2 \text{ min}\cdot\text{day}^{-1}$ or $\sim 218 \text{ min}\cdot\text{week}^{-1}$ in low-fit and $51.8 \pm 31.4 \text{ min}\cdot\text{day}^{-1}$ or $\sim 363 \text{ min}\cdot\text{week}^{-1}$ in high-fit) approached the recommended target of $150 \text{ min}\cdot\text{week}^{-1}$ (Garber et al., 2011). High-fit women were $\sim 1 \text{ h}\cdot\text{day}^{-1}$ less sedentary ($P < 0.01$), they recorded on average 3156 more daily steps ($P < 0.01$), and they engaged in twice as much time ($P < 0.01$) at absolute MVPA compared to low-fit women. However, when PA intensity boundaries were individually

tailored, daily time spent at MVPA₊₁₀ was significantly reduced ($P < 0.01$) by ~40% in low-fit and by ~60% in high-fit groups. These data revealed no differences ($P = 0.62$) between groups (~20 min·day⁻¹ in 10-min). Accordingly, the number of participants meeting PA guidelines dropped from 62% to 32% in low-fit and from 82% to 40% in high-fit ($P < 0.01$). The only difference between groups using relative intensity cut-points was in low-light PA. The high-fit group spent ~1 h·day⁻¹ more at the lowest intensity activities ($P < 0.01$).

*** Table 1

Discussion

In this study, we tested whether individually-tailored cut-points based on lactate thresholds differed in the time spent at different PA intensity levels to fixed cut-points obtained at absolute moderate intensity (*i.e.*, 3–6 METs) (Sasaki et al., 2011) in postmenopausal women differing in CRF. Our findings demonstrated that high-fit women were 1h less sedentary, recorded ~3000 more daily steps and twice as much time at MVPA compared to low-fit individuals using absolute intensity cut-points. However, when PA intensity cut-points were individually tailored according to LT, the time spent at MVPA was significantly reduced by 60% in high-fit and by 40% in low-fit groups, showing no difference in the time spent at MVPA between groups.

Fixed or individually tailored cut-points. The role of cardiorespiratory fitness

The selection of appropriate accelerometer cut-points to demarcate PA intensity levels is the cornerstone to obtain reliable PA outcomes and their association with health markers and mortality (Rejeski et al., 2015). LT represent individual physiologic adaptations in the use of energy pathways, and they are accurate indicators of relative exercise intensity (Binder et al., 2008). The average activity counts at LT1 were 4133 ± 1152 ct·min⁻¹, showing a substantial interindividual variability (~2000–10000 ct·min⁻¹) due to the large range in LT1 (3.6–7.3 km·h⁻¹

¹). This value is 54% and 29% higher than previously reported values of 2690 (Sasaki et al., 2011) and 3208 $\text{ct}\cdot\text{min}^{-1}$ (Santos-Lozano et al., 2013) for the lower boundary of moderate intensity (3 METs), respectively. These differences could be related to the higher energy expenditure value estimated at LT1 (3.7 ± 0.7 METs) (Weyand et al., 2013) compared to the classic 3 METs threshold. Besides, the above mentioned studies (Santos-Lozano et al., 2013; Sasaki et al., 2011) obtained ~ 4000 $\text{ct}\cdot\text{min}^{-1}$ at $4.8 \text{ km}\cdot\text{h}^{-1}$, which is closer to the value of 3869 $\text{ct}\cdot\text{min}^{-1}$ found in our study at the same speed, suggesting that the 3 METs classic boundary falls below the LT1, and thus is not representative of the lower relative moderate intensity boundary in our group of postmenopausal women.

The activity counts measured at LT2 (6783 ± 2077 $\text{ct}\cdot\text{min}^{-1}$) were 9% higher and 21% lower than those reported by Sasaki et al. (2011) and by Santos Lozano et al. (2013), respectively, at the upper classic boundary of moderate intensity (6 METs). These two studies used an incremental treadmill protocol with only 2-4 stages to derive activity counts cut-points at 3 and 6 METs. In contrast, our IST provided a minimum of seven data points from all participants, with small speed increments ($0.6 \text{ km}\cdot\text{h}^{-1}$). Besides, we directly determined the activity counts at LT1 and we used individual, rather than whole-group regression equations to determine the activity counts at LT2. Our results go hand-in-hand with the findings of Ozemek and colleagues (2013) showing that CRF (*i.e.*, LT2 velocity) ($R^2=0.35-0.56$), rather than age or body mass index ($R^2 < 0.03$) influenced individual's activity count cut-points at relative intensities (Figure 3). These findings are evidence that individually tailored cut-points may better represent intensity-specific PA levels.

Implications in the measurement of physical activity

Large variations exist in the volume of objectively monitored MVPA across different countries and studies, and, therefore, in the percentage of adults meeting current PA

recommendations (Cerin et al., 2014; Colley et al., 2011; Kujala et al., 2017; Tucker et al., 2011). These differences could be explained in part by the non-harmonized data collection, and the use of different activity monitors and intensity cut-points between studies (Miguelles et al., 2017). In our study, using the absolute intensity approach, daily time spent at MVPA₊₁₀ was 42 min·day⁻¹ or 294 min·week⁻¹, and 72% of the study participants met the MVPA target. These values are higher than the 8 min·day⁻¹ (Tucker et al., 2011) or the 21 min·day⁻¹ (Colley et al., 2011) reported by US and Canadian women, corresponding to 7% and 15% of American and Canadian women meeting PA guidelines, respectively. The high number of daily steps (~12300) also confirms that our sample was more active than the vast majority of women in previous studies (Colley et al., 2011; Tucker et al., 2011). This finding is supported by the results of the IPEN study (Cerin et al., 2014), where, among the 17 city-regions, the participants from Pamplona (Spain) recorded the highest levels of MVPA (51 min·day⁻¹ in 1-min bouts). Our findings strongly suggest that CRF, rather than BMI has a stronger relationship with both, MVPA and daily steps ($P < 0.01$, $r = 0.49$ and 0.53 vs. $r = -0.24$ and -0.28 , respectively), without being affected after adjusting for BMI or age. An important finding of our study was that MVPA was significantly reduced ($P < 0.01$) by half (40% in low-fit and 60% in high-fit) when the cut-points were individually-tailored. Thus, the data confirmed that the use of fixed absolute intensity cut-points derived from 3 and 6 MET intensities overestimated MVPA.

Another important finding of this study was that although the high-fit group was ~1h less sedentary, recorded ~3000 more daily steps and twice as much time at absolute MVPA compared to the low-fit group, these differences disappeared when PA levels were analysed according to their relative intensity cut-points, and both groups recorded similar time at MVPA (~20 min·day⁻¹ in 10-min bouts). Our results are in line with a large-scale Finish study (Kujala et al., 2017) who found that using HR-based absolute intensity thresholds men recorded twice as much MVPA than women (~45 vs ~22 min·day⁻¹ in 1-min bouts). However, using relative

intensity thresholds, MVPA was reduced by half ($\sim 17 \text{ min}\cdot\text{day}^{-1}$) without differences between them. Overall, this information is evidence that using fixed accelerometer activity cut-points derived from the recommended 3-6 METs moderate PA intensity approach may not be the most accurate option when examining PA behavior in samples varying in CRF.

Strengths and limitations of the study

This is the first study measuring accelerometer-based PA using individually tailored cut-points corresponding to LT. Despite the methodological shortcomings of accelerometers-based measures in PA surveillance (Pedisic & Bauman, 2015), the proposed method in this study avoids the misinterpretation of intensity-specific PA levels when using fixed activity cut-points in individuals or groups varying in CRF. However, the current study is not without limitations. First, the applicability of this individualized approach is not feasible for large epidemiological studies, although they could benefit from population-specific relative intensity cut-points to obtain more reliable PA outcomes. Second, the generalizability of these findings is limited to postmenopausal women, who are worthy of distinct attention due to the increased risk of cardiovascular disease (Kannel et al., 1976) and osteoporosis (Whitfield et al., 2015). Third, our study was a cross-sectional study. Although higher relative intensity of daily activities has been associated with reduced CHD mortality (Lee et al., 2003), randomized controlled trials are needed to confirm whether PA recommended by guidelines applying relative intensity thresholds brings greater long-term health benefits compared to absolute intensity. Fourth, accelerometers do not capture accurate energy expenditure of certain activities (*e.g.*, cycling, swimming, weight training, and some complex low-intensity activities) (Matthew, 2005). Besides, habitual PA could be much more variable than constant velocity exercise tests used to demarcate PA intensity categories, in a way that makes the same $\text{ct}\cdot\text{min}^{-1}$ more intense (*e.g.*, uphill, involving arm movement or resistance). Nonetheless, LT are the gold standard markers

of relative exercise intensity, and the present focus on walking exercise tests is appropriate, because it is the most common type of activity among adults (Arem et al., 2015).

Conclusion

This study demonstrated that applying recommended absolute moderate intensity criteria of 3-6 METs to accelerometer activity counts cut-points overestimated the time spent at MVPA. Compared to low-fit individuals, it was easier for high-fit individuals to reach MVPA target according to absolute criteria. However, no differences were observed in the time spent at MVPA between high- and low-fit postmenopausal women when the intensity cut-points were individually tailored to their lactate thresholds velocities. Our findings suggest that individually-tailored accelerometer cut-points may provide a more representative PA profile of individuals differing in CRF, compared to the widely used absolute intensity cut-points.

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Conflicts of interest and source of funding:

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Table 1. Comparison of daily physical activity levels ($\text{min}\cdot\text{day}^{-1}$) according to relative or absolute intensity activity cut-points in high and low cardiorespiratory fitness groups.

	Low-fit (n=37)		High-fit (n=38)	
Age (years)	61.4 ± 5.8		56.6 ± 4.0**	
Height (cm)	157.8 ± 5.0		159.3 ± 5.8	
Body mass (kg)	69.7 ± 12.7		61.4 ± 6.0**	
BMI (kg/m^2)	27.9 ± 4.3		24.2 ± 2.5**	
LT1 ($\text{km}\cdot\text{h}^{-1}$)	4.7 ± 0.5		5.5 ± 0.6**	
LT2 ($\text{km}\cdot\text{h}^{-1}$)	6.3 ± 0.4		7.8 ± 0.9**	
Steps	10682.1 ± 3402.0		13837.7 ± 4074.3**	
Sedentary ₊₁ ($\text{min}\cdot\text{day}^{-1}$)	503.2 ± 103.7		449.9 ± 96.7*	
Sedentary ₊₁₀ ($\text{min}\cdot\text{day}^{-1}$)	426.0 ± 121.7		363.4 ± 104.7*	
<i>Activity intensity level ($\text{min}\cdot\text{day}^{-1}$)</i>	<i>Relative</i>	<i>Absolute</i>	<i>Relative</i>	<i>Absolute</i>
Low-light	342.6 ± 86.2	297.6 ± 69.5††	402.9 ± 78.8 [‡]	311.5 ± 65.9††
High-light	75.8 ± 34.0	99.1 ± 39.5††	73.9 ± 27.6	112.7 ± 32.5††
Low-mod	20.6 ± 16.5	41.8 ± 19.5††	25.2 ± 26.7	47.9 ± 18.9††
High-mod	7.5 ± 11.4	13.2 ± 18.5 [†]	6.2 ± 8.3	31.1 ± 24.2*††
Vigorous	3.3 ± 7.8	0.7 ± 1.5††	2.3 ± 5.2	6.0 ± 8.9*††
MVPA ₊₁	31.4 ± 24.7	55.6 ± 28.0††	33.7 ± 28.5	85.0 ± 32.9*††
MVPA ₊₁₀	18.8 ± 21.2	31.1 ± 24.2††	21.6 ± 25.9	51.8 ± 31.4*††
<i>Meeting PA guidelines (%)</i>	32.4	62.2††	39.5	81.6 [†]

Data presented as mean ± standard deviation or percentages. Low-fit = Low LT2 velocity group, High-fit = High LT2 velocity group, relative = relative intensity method based on lactate thresholds, Absolute = absolute intensity method based on measured activity counts at 3-6 METs (Sasaki et al., 2011), MVPA = Moderate-to-vigorous physical activity. Physical activity (PA) guidelines = $\geq 150 \text{ min}\cdot\text{wk}^{-1}$ of MVPA in 10-min bouts (Garber et al., 2011). Sedentary time and MVPA are reported in 1 and 10 minute bouts (+1, +10, respectively).

† Significant intra-group differences between relative and absolute cut-points ($P < 0.05$).

†† Significant intra-group differences between relative and absolute cut-points ($P < 0.01$).

* Significant inter-group differences at absolute cut-points ($P < 0.05$).

** Significant inter-group differences at absolute cut-points ($P < 0.01$).

[‡] Significant inter-group differences at relative cut-points ($P < 0.05$).

Figure captions

Figure 1. **a)** Schematic representation of the 20 m track used in the IST and CVT. **b)** Blood lactate and heart rate response during the IST for the individual determination of the LT1 in a representative participant. The participant started running at 6.7 km·h⁻¹ **c)** Lactate response during several CVT for the determination of the LT2 in the same representative participant. Three metabolic intensity zones: light (<4.25 km·h⁻¹), moderate (4.25-7.45 km·h⁻¹) and vigorous (>7.45 km·h⁻¹) are indicated by different background colours (green, yellow and red, respectively). LT1 = first or aerobic threshold, LT2 = second or anaerobic threshold, W: walk, R: run.

Figure 2. Blood lactate concentration and accelerometer activity counts during the IST. Data are mean values for the whole sample (n = 75). Five metabolic intensity zones: low-light and high light (<LT1), mod-low and high-low (between LT1 and LT2) and vigorous (>LT2) are indicated by different background colours (blue-green, yellow-orange and red, respectively). [La⁻] = blood lactate, ct·min⁻¹ = activity counts per minute, LT1 = first or aerobic threshold, LT2 = second or anaerobic threshold, V_{max} = maximal velocity obtained during the test, VM₃ = vector magnitude activity counts, MET = estimated metabolic equivalents (Weyand et al., 2013), W, W&R, R = number of subjects walking, a combination of walking and running or running, respectively, at each stage. * Significantly (P <0.01) different from the previous stage.

Figure 3. Tukey Box Plot showing the median and the 25-75 percentiles of the VM₃ activity counts at LT1 and LT2 velocities in Low-fit (red coloured) and High-fit (blue coloured) groups. The grey coloured area shows the VM₃ activity counts cut-points derived from walking and running activities that represent an intensity of 3 and 6 METs (Sasaki et al., 2011). VM₃ = vector magnitude, MET = metabolic equivalent, LT1 = first or aerobic threshold, LT2

= second or anaerobic threshold, Low-fit = low aerobic fitness group ($\leq 6.8 \text{ km}\cdot\text{h}^{-1}$), High-fit
= High aerobic fitness group ($> 6.8 \text{ km}\cdot\text{h}^{-1}$). * Significantly different between groups ($P < 0.01$).