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Reliability of peak O₂ uptake and O₂ uptake kinetics in step exercise tests in healthy subjects



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ABSTRACT

To date little is known about the reliability of peak oxygen consumption ($\dot{V}O_{2PEAK}$) in incremental metronome paced step tests (IST) and the reliability of on-kinetics $\dot{V}O_2$ has never been studied. We aimed to study the reliability of both tests. Eleven healthy subjects performed two ISTs until exhaustion. On two different days two duplicate 4 min constant metronome paced step tests (CST) were performed. $\dot{V}O_{2PEAK}$, mean response time (MRT) and phase II time constant (τ) were tested for reproducibility using the paired t-tests, in addition to the limits of agreement (LOA) and within subject coefficient of variation (COV). With a 95% LOA of 0.38 to 0.26 L min $^{-1}$, -8.7 to 9.1 s and -9.9 to 10.5 s they exhibit a COV of 3%, 4.5% and 6.9% for $\dot{V}O_{2PEAK}$, MRT and τ respectively. ST are sufficiently reliable for maximal and submaximal aerobic power assessments in healthy subjects and new studies of oxygen uptake kinetics in selected patient groups are warranted.

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1. Introduction

Step tests (ST) elicit vigorous exercise intensities in healthy subjects (Hansen et al., 2011; Petrella et al., 2001; Turner et al., 2004) or selected pulmonary patients (Dal Corso et al., 2013) and are actually increasingly performed in cardiopulmonary research and medical practice (Dal Corso et al., 2007; de Camargo et al., 2011; Fox et al., 2013; Woods et al., 2012).

The practical advance of ST is to allow measurement of the valid cardiopulmonary response without requiring expensive ergometer equipment. However, it has been underutilized and there is a major gap in its applicability due to the lack of well-defined protocols and scarce knowledge about ST reproducibility for measuring aerobic power (Petrella et al., 2001), including $\dot{V}O_2$ kinetics. Such analysis is important in order to determine the effects of training and rehabilitation (Jones and Burnley, 2009), heart transplantation

(Jendzjowsky et al., 2007) and pharmacological effects (Berton et al., 2010), for example.

Studies on the reproducibility of $\dot{V}O_2$ at peak exercise on a cycle ergometer show a coefficient of variation (COV) of generally below 10% (Akkerman et al., 2010) and the kinetics of $\dot{V}O_2$ showed good results in terms of reproducibility in healthy subjects (Markovitz et al., 2004; Kilding et al., 2005). However, the reproducibility of $\dot{V}O_2$ at peak in ST is discordant, although, in general, it has shown good results in earlier studies (Jones et al., 1987; Siconolfi et al., 1985). Thus, knowing the reliability of the ST in the evaluation of $\dot{V}O_{2PEAK}$ and testing its usefulness in measuring the initial kinetics are key factors in proposing it as an alternative modality in exercise physiology.

Because the ST is a test that faithfully reproduces more activities of daily living, such as climbing stairs or a slope (as treadmill), we propose, in this study, to research, for the first time as far as we can ascertain from a search in the literature, the validity of the ST for the on- $\dot{V}O_2$ kinetics. We hypothesized, that the $\dot{V}O_2$ at peak exercise and the parameters of on-kinetics in the ST are reproducible and easily applied in healthy subjects with the chance of becoming a valid and applicable method for future clinical studies.

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2. Material and methods

2.1. Participants

Fourteen non-trained, healthy individuals, recruited from our University Hospital, volunteered for the study. All participants met the following inclusion criteria: aged between 18 and 60 years at entry, not addicted to alcohol or tobacco and without any known disease or current treatment. Participants with joint or musculoskeletal limitations, obesity (even grade I), hypertension, or any other pathology or taking any medication were not included in the study. The consent form was signed by the subjects after an extensive and detailed explanation of the techniques to be applied. After physical examination, all subjects attended on four occasions, over the period of one week, in order to complete the tests. The study was approved by the ethics committee on human research of our institution and is in accordance with the declaration of Helsinki.

2.2. Study design

In this test and retest reproducibility study, we initially conducted a pilot study in which several individuals underwent ST to estimate the rate of step increase and the best exercise time interval for the constant cadence step test (4 or 6 min). On the subjects' first visit, a physical examination and detailed medical history was performed, in addition to a thorough explanation of all the tests. The subjects were scheduled to perform the exercise tests on four different days, over a period of one week. On the first and third visits identical incremental tests were performed at the same time of day (± 30 min). On the second and fourth visits two identical tests were applied each day, with a constant cadence, for reproducibility of the kinetics of $\dot{V}O_2$. In all tests (IST or CST) the examiners were blinded to previous step test results. Subjects were instructed not to change their physical activity profile during the week of testing, abstain from coffee, chocolate, tea or other stimulants and consume a light meal 2 h before testing.

2.3. Measurements

2.3.1. Step testing protocol

The ST was based on the assumptions of the 6-minute walk test of the American Thoracic Society/American College of Chest Physicians (Laboratories, 2002) and on a previous ST study (Dal Corso et al., 2007), standardized by the examiner with incentives every minute (e.g., "you are doing well, continue.") on a 20 cm high step without support.

On the first and third visits a symptom-limited incremental ST (IST) was conducted driven by a signal emitted by a metronome, starting at 106 beats min⁻¹ with increments of two beats every 20 s. Each beat began on a movement of a leg up or down. The test began with the collection of measurements in the sitting position (i.e., measurement of blood pressure (BP), heart rate (HR) and peripheral oximetry (Dixtal DX 2010, São Paulo, Brazil) at rest. Upon assuming the standing position, the participants proceeded with the following test phases: (i) baseline (2 min), (ii) exercise until exhaustion and (iii) recovery (2 min). BP, HR, oximetry and the rate of perceived exertion (RPE), using the 20-point Borg scale (Borg, 1982), were collected at the end of the exercise.

On the first visit (IST) the detection of the cadence (beats min⁻¹) at peak exercise (i.e., when the subject could not sustain the exercise for more than 10 s) was planned, sufficient to induce subsequent levels of exercise in the constant cadence step test (CST) which was set at 80–90% of maximum cadence. We assumed that there was a load applied to move up or down which was linearly

related to speed (cadence) during the ST, similar to the concept defined by Porszasz et al. (2003) in a study on the treadmill:

$$S \times t = m \times g \times (V_s \times t) \times f(t) \tag{1}$$

where, S is the slope work rate (W min⁻¹), m = mass, g = gravitational constant, V is the rate of change in speed (rate in ms⁻²) and f is the time course of sin (α), in our case, a constant value, unlike the treadmill exercise, where there is an elevation change. Therefore, the load or the power (Eq. (1)) is directly proportional to speed, and if the speed is increased at a set time (every 20 s), the test is a test for incremental setting. The \dot{V} O_{2PEAK} criterion was based on observation of maximum exertion (which could not be sustained for more than 10 s) and a respiratory quotient above 1.1.

The second and fourth visits were designated for CST, obtained with the cadence calculated as 80-90% of the maximum rate of the first IST. The tests were duplicated with an interval of 30 min of rest on each day of testing. In a quiet environment, with only the examiner, an assistant and the subject, the tests, which were preceded by a rigorous and comprehensive explanation of all procedures, were conducted in the following stages, after measurement of vital signs and resting RPE: (i) baseline (3 min, standing), (ii) constant paced exercise for 4 min and (iii) recovery (sitting for 5 min). The same variables of IST were taken at the beginning and end of the period. We chose 4 min and not 6 min for three reasons: (i) previous study have shown that 3 or 4 time constant (TC, i.e., the time when 63% of steady state $\dot{V}O_2$ value is achieved) is sufficient to determine the fundamental TC (Bell et al., 2001), (ii) an earlier study had indicated that a plateau was obtained at a speed similar to ours in less than 3 min (Jones et al., 1987) and (iii) in the pilot study we observed, after 3 to 4 min, a higher frequency of fatigue and exercise termination, coupled with the fact that up to 12% of the ST individuals in a previous study did not complete 5 min of exercise because of physical exhaustion (Hansen et al., 2011). It was not our goal to establish a level of exercise above or below lactate threshold, but to establish a high speed (80-90% of maximum cadence), in order to optimize the amplitude of the $\dot{V}O_2$, provided care is taken to verify the absence of "slow component" within 3 min (Chiappa et al., 2008; Laveneziana et al., 2009; Berton et al., 2010; Vasilopoulou et al., 2012) and taking into account that the fundamental TC does not change significantly with exercise above or below the lactate threshold (Özyener et al., 2001).

2.3.2. Metabolic data collection

The exercise tests were performed in the metabolic system model Vmax 229 (Vyasis, USA), breath by breath, calibrated before each test with precision gases (Gases Gama, São Paulo, Brazil) for two reference points ((i) 26% O₂ and nitrogen balance, (ii) 16% O₂ and 4% CO₂, balanced with nitrogen). Measurement of inspiratory and expiratory gas flow was performed through a bidirectional, low dead space (39 mL) and low resistance ($<1.5 \text{ cmH}_2\text{O/L/s}$ at 12 L s^{-1}) mass flow sensor (Viasys, Yorba Linda, CA), calibrated before each test with a 3 L syringe and attached to a face mask (Hans Rudolph, Kansas City, MO., USA). The signals from the electrochemical cell analyzer and mass flow sensor were interfaced to the computer via analog-digital integrated dedicated software (Vyasis, Yorba Linda, CA) and the results exported to an excel spreadsheet. Individuals were monitored continuously using a 12-lead ECG (Cardiosoft, SensorMedics, Yorba Linda, CA). Through this system we obtained breath by breath $\dot{V}O_2$, rate of carbon dioxide production ($\dot{V}CO_2$), pulmonary ventilation per minute ($\dot{V}_{\rm E}$) and HR.

2.3.3. Cardiopulmonary data analysis

To calculate the $\dot{V}O_{2PEAK}$ the 10 s average of all measures breath by breath in the incremental test was obtained until the criterion of maximum effort and the greatest $\dot{V}O_2$ achieved in the last minute

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