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Determination of anaerobic threshold through heart rate and near infrared spectroscopy in elderly healthy men

Michel S. Reis1, Danilo C. Berton2, Ross Arena3, Aparecida M. Catai4, José A. Neder2, Audrey Borghi-Silva4

ABSTRACT | Background: Aging leads to low functional capacity and this can be reversed by safe and adequate exercise prescription. Objective: The aim of this study was to identify the anaerobic threshold (AT) obtained from the V-slope method as well as visual inspection of oxyhemoglobin (O₂Hb) and deoxyhemoglobin (HHb) curves and compare findings with the heteroscedastic (HS) method applied to carbon dioxide production (V CO₂), heart rate (HR), and HHb data in healthy elderly men. A secondary aim was to assess the degree of agreement between methods for AT determination. Method: Fourteen healthy men (61.4±6.3 years) underwent cardiopulmonary exercise testing (CPX) on a cycle ergometer until physical exhaustion. Biological signals collected during CPX included: ventilatory and metabolic variables; spectroscopy quasi-infrared rays – NIRS; and HR through a cardio-frequency meter. Results: We observed temporal equivalence and similar values of power (W), absolute oxygen consumption (VO₂ - mL/min), relative VO₂ (mL.Kg⁻¹.min⁻¹), and HR at AT by the detection methods performed. In addition, by the Bland-Altman plot, HR confirmed good agreement between the methods with biases between –1.3 and 3.5 beats per minute. Conclusions: (i) all detection methods were sensitive in identifying AT, including the HS applied to HR and (ii) the methods showed a good correlation in the identification of AT. Thus, these results support HR as valid and readily available parameter in determining AT in healthy elderly men.

Keywords: physical therapy; anaerobic threshold; NIRS; cardiopulmonary test; heteroscedastic model.

Introduction

Aging is associated with changes in the structure and function of the cardiovascular, pulmonary, and muscle systems. These adaptive processes lead to low functional capacity and perpetuate a sedentary lifestyle. In this context, it seems rational to devise reliable strategies of exercise prescription for sedentary elderly.

The integration of the cardiovascular, respiratory, and muscular systems constitutes a complex and sophisticated mechanism of energy generation during physical exertion. Anaerobic threshold (AT or AT1) has been an important index of performance for assessing cardiopulmonary and muscle integration during aerobic exercise. Additionally, the respiratory compensation threshold (RCT or AT2) has been useful for determining the attainment of quasi-maximum intensities.

The analysis of ventilatory and metabolic responses obtained during progressive cardiopulmonary exercise testing (CPX) reflects the cellular respiration of peripheral muscles; however, it occurs with some delay. This delay may be explained by a slowdown in the gases circuit filling all the volume available in its muscle and alveolar regions, which is measured at the mouth, as well as factors associated with oxygen and carbon dioxide diffusibility.

Recent studies have shown that the identification of metabolic transition phases (AT1 and AT2) can be observed by the dynamic assessment of relative concentrations of oxyhemoglobin (O₂Hb) and deoxyhemoglobin (HHb) through near infrared spectroscopy. During incremental exercise testing, O₂Hb presents two sequential decreasing patterns according to exercise intensity: a major deflection,
which coincides with AT1, followed by a slower deflection, related to AT2. On the other hand, although most studies have assessed only O2Hb behavior, HHb seems to demonstrate the same sequential patterns (i.e. almost a mirroring behavior), but with an increasing trajectory.

Another tool which has been useful for determining thresholds is the heart rate (HR) response to progressive aerobic exercise. Previous laboratory studies show that mathematical models applied to carbon dioxide production (VCO2) data and root mean square (RMS) of electromyographic signal demonstrate a strong relationship with HR data in apparently healthy subjects. In this context, using the heteroscedastic mathematical model applied to data, Pozzi et al. asserted that VCO2, HR, and RMS present a strong relationship with graphic visual analysis of ventilatory parameters, still considered the gold standard in AT detection.

With this in mind, we hypothesize that AT1 determination in elderly healthy men through ventilatory and metabolic variables, O2Hb and HHb curves, and HR behavior may reveal temporal similarities between the methods. Additionally, according to interdependent physiological events, the methods could represent a simpler and more affordable strategy. Therefore, the primary aim of this study was to identify AT1 obtained through the gold standard method, O2Hb and HHb visual method, and compare it to the heteroscedastic mathematical model applied to VCO2, HHb, and HR data. Secondly, we aimed to evaluate the level of agreement amongst the methods for determining AT1. Finally, the knowledge of new strategies for exercise prescription in the elderly is important to physical therapy. Safe and appropriate tools for individualized exercise prescription will ensure a successful approach to improve functional capacity and quality of life in aging.

• Method

Experimental approach to the problem

This investigation required subjects to perform a CPX on a cycle ergometer until physical exhaustion (in ramp protocol). During the exercise effort, ventilatory and metabolic, NIRS, and HR variables were simultaneously recorded. Additionally, the investigators applied the heteroscedastic model to the datasets to identify the AT and compared it to the moment of AT by the visual method (gold standard method for AT determination). In this sense, these outcomes could be used subsequently to establish the viability of the additional methods for evaluating the AT during aerobic conditioning programs.

Subjects

Fourteen healthy individuals were recruited through clinical assessment (mean±SD age, height, weight, and body mass index=61.4±6.3 years, 1.71±0.05 m, 75±6.3 Kg, and 24.3±3.0, respectively). All subjects presented with: a good general health status; absence of cardiovascular, respiratory, or musculoskeletal system abnormalities, as well as absence of metabolic alterations; and diminished aerobic capacity consistent with a sedentary lifestyle, according to CPX results. None of the subjects were: tobacco users, alcohol-dependent, or users of addicting drugs; diagnosed with diabetes mellitus, dyslipidemia or systemic arterial hypertension; or taking prescribed antihypertensive or cardioactive drugs.

All subjects were submitted to: a clinical assessment (current and past clinical history, family background, life habits, physical exams) with a pneumologist; a physiotherapeutic assessment (postural assessment and muscle tests); dyspnea evaluation; laboratory exams (complete blood count, triglyceride levels, total and fraction cholesterol, urine type I, uric acid, creatinine, and urea); spirometry; 12-lead electrocardiography (ECG); and maximal CPX. We excluded all individuals who presented ECG alterations that would not allow the determination of heart rate variability (HRV); and who did not meet the inclusion criteria listed above.

The subjects were informed on the experimental procedures to which they would be submitted and signed the informed consent form before taking part in the study. Approval was granted by the Ethics Committee of Universidade Federal de São Paulo (UNIFESP), São Paulo, SP, Brazil (084/06).

Procedures

Data collection was carried out in an air-conditioned laboratory at 22 °C to 24 °C and 50 to 60% relative humidity in the same period of day (between 8 a.m. and 12 p.m.). The subjects were familiarized with the experimental environment and research personnel. The day before and on the day of the test itself, each subject was instructed to: avoid stimulating drinks; refrain from physical exercise 24 hours prior to data collection; have a light meal in the morning of data collection; and have an adequate period of sleep (at least 8 hours). CPX was carried out with a ramp protocol on an electromagnetic braking cycle ergometer (Corival, Lode BV, Groningen, The Netherlands) in the upright sitting position. Initially, subjects had 2 minutes of rest sitting on the
cycle ergometer; subsequently, a 3-minute warm up period began, pedaling without load (about 4 W). After this stage, the exercise protocol started with power increments of W/min (determined according to the functional capacity reported by subjects during clinical assessment – 15-25 W) and constant speed of 60 rpm until physical exhaustion – i.e., inability to maintain predetermined pedaling speed. The load distribution was controlled through the ventilatory expired gas analysis system (CardiO₂ System, Medical Graphics Corporation, St. Paul, MO, USA). The post-test recovery monitoring period involved 3 minutes of active recovery followed by 2 minutes of rest. Ventilatory and metabolic variables were collected throughout the CPX. Pulse oximetry (SpO₂; Minolta, Stowwood Scientific Instruments, Oxford, UK) and ECG (CardiO₂ System, Medical Graphics Corporation, St. Paul, MO, USA) – in derivations MC5, DII, DIII, aVR, aVL, and aVF modified and V1 to V6 – were continually monitored during all experimental procedures and arterial pressure was intermittently assessed throughout the protocol. The tests were carried out by a team of researchers, composed of physical therapists and physicians, who monitored physiologic responses and signs/symptoms exhibited by subjects.

**Pulmonary function**

Pulmonary function testing, measuring slow vital capacity (SVC), forced vital capacity (FVC), forced expiratory volume in one second (FEV₁), and FEV₁/FVC ratio, was carried out through the CardiO₂ System (Medical Graphics Corporation, St. Paul, MO, USA). For comparative purposes, reference values from Knudson et al.¹⁶, expressed in body temperature pressure standard (BTPS) conditions, were used. Carbon monoxide diffusion capacity (D CO) was assessed through the simple respiration model and static volumes were assessed through whole body plethysmography. Technical procedures and the acceptability and reproducibility criteria were defined according to guidelines recommended by the American Thoracic Society¹⁷.

**Ventilatory and metabolic variables.**

The ventilatory and metabolic variables were obtained through a computer-aided ergospirometric measurement system (CardiO₂ System, Medical Graphics Corporation, St. Paul, MO, USA) using the Breeze Suite 6 software package. Tidal volume was obtained through a Pitot pneumotachometer connected to the CardiO₂ System and attached to a face mask selected according to the subject’s size and providing an adequate fit in order to avoid air leakage. The device presents in real time applied power values (W), pedaling speed (rpm), as well as oxygen consumption (VO₂) and carbon dioxide production (VCO₂), minute ventilation (VE), HR, and SpO₂. Ventilatory equivalent values (VE/VO₂ and VE/VCO₂), respiratory exchange ratio (RER), end-tidal partial pressure of oxygen (PETO₂), and carbon dioxide (PETO₂), flow-volume (FV), and respiratory rate (FR) were also calculated and recorded. The power applied to the cycle ergometer during exercise protocols was controlled by the system through an interface with the bicycle.

**Near Infrared Spectroscopy (NIRS)**

This methodology was carried out through NIRS (NIRO 300, Hamamatsu Photonics, Hamamatsu, Japan) at a wavelength between 775 and 905 nm and a distance of 3 cm between the generator and detector, allowing the light to penetrate tissue ~ 1.5 cm. Subsequently, the detection of light attenuation on small caliber vessels allows determination of relative concentrations of O₂Hb, HHb, and total hemoglobin. This computerized system has two channels that allowed us to position the optodes (containing light generator and detector) on the vastus lateralis – about 12-14 cm above the knee. The optodes were tightly positioned in order to avoid movement during exercise. They were also protected from environment light, avoiding its interference in data collection. Data were recorded and stored in the system each second and the protocol transition points were accurately marked to allow for an adequate data analysis.

**Heart rate and RR intervals**

HR and RR intervals (RRi) were recorded beat by beat through a cardio-frequency meter (Polar® S810i) with a 1,000 Hz sampling frequency fastened by an elastic band to the lower third of the sternum, providing simultaneous transmission to the watch where data were stored. Afterwards, through a serial port interface with an infrared sensor, data were transferred and stored in a personal computer (Pentium III, 1,100 MHz) to be analyzed. The protocol transition points were also accurately marked to allow for an adequate data analysis.

**Analysis methodology**

**Visual ventilatory method**

Visual analysis of VO₂ and VCO₂ correlation curves, the VE/VO₂ and VE/VCO₂ ratios, and PETO₂
and \( P_{ET} CO_2 \) were graphically represented in moving mean values every 8 respiratory cycles. Subsequently, three independent observers determined AT under the following situations\(^{19,20}\): 1) \( V \)-slope: breaking point from linearity in the \( VO_2 \) and \( VCO_2 \)-correlation curves; 2) \( VE/VCO_2 \): nadir point of this ratio, ensuring that from it a systematic increase occurs; and \( P_{ET} O_2 \): nadir point of this variable, from which a systematic increase begins.

The section selected for subsequent AT determination was identical for both methods. It was set from the beginning of the responses of ventilatory and metabolic variables to power output increments to the end of exercise. The analysis of each observer was performed in an independent manner, on a 15 inch monitor (SyncMaster 550V, Samsung) connected to the MedGraphics software. Power (W), \( VO_2 \) (mL/min), \( VO_2 \) (mL.Kg\(^{-1}\).min\(^{-1}\)), and HR values were obtained and calculated on the variables data spreadsheet created by the ergospirometry system, interpolated second by second.

The qualitative control of experiment was performed according to many criteria: constant rotation speed (60 rpm) until physical exhaustion; presence of artifacts which could affect the quality of test and measurement of AT; presence or absence of a balance state in the warm up period; investigation of a co-occurrence between power increment and the emergence of HR responses and ventilatory variables; and investigation of a linear behavior of ventilatory variables at the beginning of the slope. This methodology was employed as gold standard when compared to the other methods for determining AT.

In addition, we measured the respiratory compensation threshold (RCT) and this information was used as a temporal orientation to select the analysis site of other methodologies employed in the current study. Thus, the observers considered the following occurrences\(^{21}\): 1) \( VE/VCO_2 \) - starting point of the systematic increase of values in this ratio; and 2) \( P_{ET} CO_2 \) - point of higher value of this variable, from which a systematic decrease starts.

**Visual method of \( O_2 \)Hb and HHb curves (NIRS)**

Visual analysis of \( O_2 \)Hb\(^a\) and HHb curves was carried out in Microsoft Excel (13.0 for Windows Student Version: For Microsoft Windows XP, 2000). Three independent observers identified the following moments: first \( O_2 \)Hb deflection and HHb inflection points at the same analytical temporal region determined in CPX. Each observer took note of the time (s) corresponding to the point identified and afterwards a three-observation mean value was calculated. From the time mean value obtained, we verified the correspondent \( VO_2 \) (mL/min), \( VO_2 \) (mL.Kg\(^{-1}\).min\(^{-1}\)), and HR capacity values in the data spreadsheet based on CPX (now interpolated each second). Visual identification of behavior change in these variables was compared to the gold standard.

**Heteroscedastic mathematical model (HS) applied to the responses of \( VCO_2 \) (HS-\( VCO_2 \), HHb (HS-HHb), and HR (HS-HR))**

The heteroscedastic model was applied through a mathematical algorithm developed using R language and environment for statistical computing (http://www.r-project.org), which determines the changing point of data series of a given variable\(^{15}\). This model was applied to HR data collected beat by beat and to \( VCO_2 \) data, respiration by respiration, and calculated in moving mean values every 8 respiratory cycles, as well as to HHb values. All data were obtained at the same analysis temporal period determined in the gold standard method. The model concerned establishes the turning point of linear data increase into an exponential increase, thereby separating two segments of the data series. This changing point in the pattern of response of these two variables defines AT. After determining AT, the data for power (W), HR (bpm), and relative and absolute \( VO_2 \) (mL.Kg\(^{-1}\).min\(^{-1}\)) at AT moment were graphically plotted and compared in order to observe the equivalence between methodologies.

The heteroscedastic simple segmented regression model, considering a sequence of observations \((y_i, x_i)\), is expressed as follows (Equation 1):

\[
y_i = \begin{cases} 
\alpha_1 + \beta_1 x_i + \epsilon_{i1}, & \text{if } i = 1,..., k \\
\alpha_2 + \beta_2 x_i + \epsilon_{i2}, & \text{if } i = k + 1,..., n 
\end{cases}
\]

where \( y_i \) is the dependent variable, \( x_i \) is an independent “fixed” variable (in this case time), and \( \epsilon_{i1} \) and \( \epsilon_{i2} \) are random errors of the relation, which are independent and usually distributed with zero mean and variance \( \sigma_i^2 \). Coefficients \( \alpha_1, \beta_1, \alpha_2, \) and \( \beta_2 \) are unknown and need to be estimated.

**Statistical analysis**

We used parametric statistical tests since data presented with a normal distribution (Shapiro-Wilk test) and heterogeneous variances (Levene test). For intragroup comparisons of continuous data, we used a one-way ANOVA and Tukey’s post-hoc test. For categorical variables, we used the non-parametric Kruskal-Wallis test. Additionally, the Pearson correlation test was employed to assess the
relationship among variables (power, HR, and relative and absolute $\dot{V}O_2$) in a comparison between the methods concerned. The abovementioned analyses were carried out using SPSS software Release 10.0.1 (1999) with a significance level of $p<0.05$. In addition, we used MedCalc version 11.2.1.0 to perform the agreement analysis of methods through Bland-Altman visual statistics for HR corresponding to the methods investigated.

### Results

Table 1 shows clinical characteristics for the cohort assessed. We did not find significant differences between visual and HS methods applied to $\dot{V}CO_2$, HHb, and HR data. Figure 1 presents a comparison among the variables power (W), time (s), $\dot{V}CO_2$ (mL.Kg$^{-1}$.min$^{-1}$), and HR (bpm) in AT determined for the methods employed. There were no significant differences in these variables among the methods.

Table 2 shows the relationship among power (W), $\dot{V}CO_2$ (mL/min), $\dot{V}CO_2$ (mL.Kg$^{-1}$.min$^{-1}$) when submitted to the gold standard method and methods V-NIRS, HS-$\dot{V}CO_2$, HS-HHb, and HS-HR in AT. All correlations were significant, but only $\dot{V}CO_2$ (mL.Kg$^{-1}$.min$^{-1}$) revealed a strong correlation between V-NIRS and the gold standard. Additionally, HR (bpm) at AT showed strong correlations with the gold standard and other methods proposed (Figure 2).

### Discussion

The main results of this study revealed that AT determination in methods V-NIRS, HS-$\dot{V}CO_2$, HS-HHb, and HS-HR presented temporal similarity while considering these correlations and with the aim of ratifying the results, we performed a Bland-Altman agreement analysis. Figure 3 shows that HR values at AT in methods V-NIRS, HS-$\dot{V}CO_2$, HS-HHb, and HS-HR showed a symmetric distribution around the mean with biases of -1.3, 0.1, 3.5, and 1.1, respectively, in agreement with the gold standard.

### Table 1. Clinical characteristics of volunteers studied.

<table>
<thead>
<tr>
<th>Pulmonary function</th>
<th>Volunteers (n=14)</th>
</tr>
</thead>
<tbody>
<tr>
<td>$FEV_1$ (L)</td>
<td>3.31±0.50</td>
</tr>
<tr>
<td>$FEV_1$ (% pred)</td>
<td>99.6±9.4</td>
</tr>
<tr>
<td>FVC (% pred)</td>
<td>103.2±10.1</td>
</tr>
<tr>
<td>$FEV_1$/FVC</td>
<td>94.4±5.8</td>
</tr>
<tr>
<td>IC (% pred)</td>
<td>97.0±11.3</td>
</tr>
<tr>
<td>$D_LCO$ (% pred)</td>
<td>86.7±9.8</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Exercise peak</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Power (W)</td>
<td>137±19</td>
</tr>
<tr>
<td>Time (s)</td>
<td>551±133</td>
</tr>
<tr>
<td>$\dot{V}O_2$ (mL)</td>
<td>1.624±267</td>
</tr>
<tr>
<td>$\dot{V}O_2$ (mL/Kg/min)</td>
<td>21.8±3.4</td>
</tr>
<tr>
<td>$\dot{V}CO_2$ (mL)</td>
<td>1.979±376</td>
</tr>
<tr>
<td>RER</td>
<td>1.22±0.12</td>
</tr>
<tr>
<td>VE (L/min)</td>
<td>74±17</td>
</tr>
<tr>
<td>HR (bpm)</td>
<td>148±22</td>
</tr>
<tr>
<td>$SpO_2$ (%)</td>
<td>96±4</td>
</tr>
<tr>
<td>Dyspnea (4-10)</td>
<td></td>
</tr>
</tbody>
</table>

Values are means±SE and the category variables in median value. $FEV_1$: forced expiratory volume in 1 second; FVC: forced vital capacity; $D_LCO$: carbon monoxide diffusing capacity; $\dot{V}O_2$: oxygen consumption; $\dot{V}CO_2$: carbon dioxide production; RER: respiratory exchange ratio; VE: ventilation (L/min); HR: heart rate (bpm); $SpO_2$: peripheral oxygen saturation.

### Table 2. Pearson correlation between variables $\dot{V}O_2$ (mL/Kg/min) and absolute $\dot{V}O_2$ (mL/min) in the method used for determining the anaerobic threshold and visual ventilatory method.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Gold standard</th>
<th>r</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Power (W)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>V-NIRS</td>
<td>0.62</td>
<td>0.04</td>
<td></td>
</tr>
<tr>
<td>HS-$\dot{V}CO_2$</td>
<td>0.70</td>
<td>&lt;0.01</td>
<td></td>
</tr>
<tr>
<td>HHb</td>
<td>0.53</td>
<td>&lt;0.01</td>
<td></td>
</tr>
<tr>
<td>HR</td>
<td>0.72</td>
<td>&lt;0.01</td>
<td></td>
</tr>
<tr>
<td>$\dot{V}O_2$ (mL/Kg$^{-1}$.min$^{-1}$)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>V-NIRS</td>
<td>0.83</td>
<td>&lt;0.01</td>
<td></td>
</tr>
<tr>
<td>HS-$\dot{V}CO_2$</td>
<td>0.60</td>
<td>0.02</td>
<td></td>
</tr>
<tr>
<td>HHb</td>
<td>0.56</td>
<td>0.03</td>
<td></td>
</tr>
<tr>
<td>HR</td>
<td>0.56</td>
<td>0.03</td>
<td></td>
</tr>
<tr>
<td>$\dot{V}O_2$ (mL/min)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>V-NIRS</td>
<td>0.75</td>
<td>&lt;0.01</td>
<td></td>
</tr>
<tr>
<td>HS-$\dot{V}CO_2$</td>
<td>0.63</td>
<td>0.01</td>
<td></td>
</tr>
<tr>
<td>HHb</td>
<td>0.50</td>
<td>0.04</td>
<td></td>
</tr>
<tr>
<td>HR</td>
<td>0.54</td>
<td>0.04</td>
<td></td>
</tr>
</tbody>
</table>

Pearson correlation ($p<0.05$). W: watts; NIRS: near infrared spectroscopy; HS: heteroscedastic model; $\dot{V}CO_2$: carbon dioxide production; HHb: deoxyhemoglobin; HR: heart rate.
to the gold standard in healthy elderly men. Consequently, power (W), \( \dot{V}O_2 \) (mL/min), \( \dot{V}O_2 \) (mL.Kg\(^{-1}\).min\(^{-1}\)), and HR (bpm) values were similar in different methods employed to determine AT. Additionally, HR was the variable that confirmed most of these findings, evidencing strong correlations and a good agreement of methods.

**Population characteristics**

The subjects in this study were eutrophic, had preserved pulmonary function, and weak aerobic power (21.8 mL.Kg\(^{-1}\).min\(^{-1}\)) compatible with a sedentary lifestyle according to the American Heart Association\(^1\). Peak RER values show that subjects achieved maximal exertion during CPX, which was further confirmed by symptoms of dyspnea and lower extremity fatigue.

**Different methodologies for determining AT**

In initial studies\(^2\) carried out with the signal obtained from analysis of the extraction of oxygen from the vastus lateralis, researchers observed a significant deoxygenation at AT. Subsequently, Bhambhani et al.\(^2\) set out to verify the similarity between the point of a significant \(O_2\)Hb decrease and the occurrence of AT. These authors demonstrated a significant correlation between the AT moment and significant \(O_2\)Hb decrease in healthy individuals during CPX using a cycle ergometer. In patients with heart failure, determining AT through NIRS has also been demonstrated. Belardinelli et al.\(^2\), who assessed 7 patients with heart failure (NYHA Class II), observed a good correlation among AT values determined through the V-slope method (changing point in the correlation curve between \(\dot{V}CO_2\) and \(\dot{V}O_2\)) and the first point of subtle deflection.
of O$_2$Hb. In the study by Miura et al.\textsuperscript{5}, VO$_2$ values obtained in AT defined by V-slope of 16 patients with heart failure revealed a similarity with VO$_2$ observed in the first point of deflection in O$_2$Hb curve. Subsequently, the same study described the correlation between a second deflection in the O$_2$Hb curve and the emergence of an increase of VE/VCO$_2$ values and decrease of P$_{ET}$CO$_2$ – both markers of PCR. Additionally, Terakado et al.\textsuperscript{7} revealed a temporal proximity between PCR and the changing point in O$_2$Hb curve in 29 patients with heart failure between AT determined in gas analysis and peak exercise. In our study, special attention was given to HHb behavior, as it represents an almost mirrored response to O$_2$Hb and has an ascending behavior, similar to the cardiovascular and metabolic variables during incremental exercise – adequate to be applied to the heteroscedastic model. The results found were interesting, since an early identification of AT (when compared to the gold standard) was expected in HHb data, due to a kinetic delay in alveolar-muscular traffic\textsuperscript{1}. However, we observed a temporal similarity between visual and heteroscedastic methods applied to HHb when compared to the gold standard. Notwithstanding, the Bohr effect seems to be the most adequate explanation to HHb behavior considering that, from the point of AT, the production and concentration of lactate is increased, turning the environment more acidic and thus causing a greater release of O$_2$ to tissues during activity\textsuperscript{8}.

Other authors\textsuperscript{24} have proposed the determination of AT through HR behavior and variability, as it is a simpler and easier method. Hofmann et al.\textsuperscript{24} did not observe significant differences between the threshold detected through the HR break point and blood lactate change point of sedentary women performing exercise with a non-continuous loading protocol. Bunc et al.\textsuperscript{25} observed that AT obtained through HR did not demonstrate significant differences when compared to AT measured through the ventilatory method, lactate or from the electromyography of the vastus medialis of the thigh.
In the study by Alonso et al., who evaluated HRV behavior in healthy individuals during maximal progressive CPX, they observed that HRV, according to the SDNN (Standard Derivation of Consecutive NN intervals) index achieved significantly lower levels at rest after AT.

In previous studies in our laboratory, a Hinkley bisegmented model was developed to determine the changing point (characterizing AT) in V\textsubscript{CO}\textsubscript{2}, HR, and RMS of the electromyographic signal (EMGs) when compared to AT measured through visual analysis method of V\textsubscript{CO}\textsubscript{2} curves and V\textsubscript{O}\textsubscript{2}. Marães et al. who studied healthy young individuals using a slope incremental protocol did not observe significant differences between the methodologies employed. In the study by Pozzi et al., who compared the Hinkley’s mathematical method and the heteroscedastic model to V\textsubscript{CO}\textsubscript{2}, HR, and RMS data in healthy elderly individuals, there were no significant differences between mathematical models and graphic visual analysis. Moreover, the authors considered that the heteroscedastic model fit itself to HR data. In our study, confirming the findings of the referred authors, we observed similarity in the AT moment obtained through ventilatory graphic visual analysis and NIRS in healthy elderly men, as well as through mathematical analysis of HS model applied to ventilatory, HR, and HHb values. Temporal similarities between methodologies suggest the validity of using any of these methodologies for determining AT, at a minimum, in individuals with similar characteristics to the cohort assessed.

Although the correlations among power and relative and absolute V\textsubscript{O}\textsubscript{2} have been statistically possible, HR analysis establishes the use of different methodologies (including HR data – beat by beat) in this population. It becomes more stimulating when we observe the good level of agreement between methodologies in the Bland-Altman analysis. Finally, the results also allow us to demonstrate the possible success of HR use as a means for assessing aerobic capacity in healthy elderly men. In this context, the...
detection of AT, which is important for individualized exercise prescription, may be effectively achieved through HR analysis.

Some constraints of this study should be considered. Although the observers started the procedures for measuring different variables in a concomitant fashion, the development of a system which could trigger the measurement of all variables at the same moment would be more appropriate in terms of precision in the selection of AT. Regarding the NIRS method, although a severe control of skinfold (lower than 2 mm) was performed, fat mass in the thigh region can interfere in the signals recorded.

**Practical application**

Our results suggest a strong association and good agreement between methodologies for determining AT when compared to the gold standard in the cohort studied. Additionally, our results suggest that the HR response is equally effective in detecting AT when compared to other methods employed in healthy elderly men. Therefore, our findings are relevant for physical therapy and they support the possible use of HR as capacity means to accurately detect AT and prescribe exercise at the safest and most appropriate individualized intensity to improve functional capacity and quality of life in aging. However, the use of these tools in other populations presenting cardiorespiratory and metabolic diseases risk factors should be investigated in future studies.

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