Brazilian Journal of Medical and Biological Research (2007) 40: 501-508
ISSN 0100-879X

# Comparison of anaerobic threshold determined by visual and mathematical methods in healthy women

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## Abstract

Several methods are used to estimate anaerobic threshold (AT) during exercise. The aim of the present study was to compare AT obtained by a graphic visual method for the estimate of ventilatory and metabolic variables (gold standard), to a bi-segmental linear regression mathematical model of Hinkley's algorithm applied to heart rate (HR) and carbon dioxide output ($\dot{V}CO_2$) data. Thirteen young (24 ± 2.63 years old) and 16 postmenopausal (57 ± 4.79 years old) healthy and sedentary women were submitted to a continuous ergospirometric incremental test on an electromagnetic braking cycloergometer with 10 to 20 W/min increases until physical exhaustion. The ventilatory variables were recorded breath-to-breath and HR was obtained beat-to-beat over real time. Data were analyzed by the nonparametric Friedman test and Spearman correlation test with the level of significance set at 5%. Power output (W), HR (bpm), oxygen uptake ($\dot{V}O_2$; mL kg$^{-1}$ min$^{-1}$), $\dot{V}O_2$ (mL/min), $\dot{V}CO_2$ (mL/min), and minute ventilation ($\dot{V}E$; L/min) data observed at the AT level were similar for both methods and groups studied ($P > 0.05$). The $\dot{V}O_2$ (mL kg$^{-1}$ min$^{-1}$) data showed significant correlation ($P < 0.05$) between the gold standard method and the mathematical model when applied to HR ($r_s = 0.75$) and $\dot{V}CO_2$ ($r_s = 0.78$) data for the subjects as a whole (N = 29). The proposed mathematical method for the detection of changes in response patterns of $\dot{V}CO_2$ and HR was adequate and promising for AT detection in young and middle-aged women, representing a semi-automatic, non-invasive and objective AT measurement.

**Key words**
- Anaerobic threshold
- Incremental test
- Graphic visual method
- Mathematical model
- Hinkley's algorithm

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Research supported by CAPES, CNPq (No. 140684/2003-4) and FAPESP (No. 01/07427-2).

Received April 28, 2006
Accepted January 12, 2007

## Introduction

The level of intensity of physical exercise, when energy production by aerobic metabolism is supplemented by anaerobic metabolism, is characterized as anaerobic threshold (AT) (1,2). This physiological delimitation provides important information about the major physiological systems of the organism involved in the performance of physical activity and constitutes a most important determinant of an individual's functional aerobic capacity (1-3).

Several methods have been employed for AT determination. The interest in the identification of the critical intensity workload above which lactate accumulation occurs has a long history (4,5). Invasive methods require repeated blood lactate concentration measurements during physical exercise (6,7) while non-invasive methods are based on the analysis of changes in the response patterns of ventilatory and metabolic variables, such as V-slope (1,5) and the graphic visual method (8,9). Some studies (8,9) have used the graphic visual method for the estimate of the disproportionate increase in ventilatory and metabolic variables, during the incremental dynamic exercise, as a gold standard for the quantification of AT. Additionally, other investigators have identified AT by analysis of heart rate (HR) (3,10-13) and of its variability (14), as well as by surface electromyography analysis (15-17).

Interest in the determination of AT has increased because of the inclusion of qualitative and systematic research in health studies. Some investigators have proposed the use of mathematical and statistical models as a means to characterize responses of biological systems of the organism during physical exercise (8-10,18,19).

The recent technological advances in equipment for the determination of AT and the use of mathematical and statistical algorithms have served to facilitate, automate and/or semi-automate this procedure. Thus, mathematical and statistical algorithms have become tools destined to represent the dynamic behavior of biological variables of interest, such as the physiological change patterns of the metabolic and ventilatory variables observed when AT is reached.

The search for ever more perfected techniques for AT determination has great importance, since this parameter has been extensively used for the evaluation of aerobic capacity at submaximal levels of physical exercise and for the prescription of individual physical training for patient rehabilitation.

In view of these considerations, the purpose of the present study was to compare the AT determined by the graphic visual method for the estimate of ventilatory and metabolic variables, considered to be the gold standard, to the Hinkley (20) bi-segmental linear regression mathematical model (MMH), in healthy women.

## Material and Methods

### Subjects

Thirteen young (24 ± 2.63 years of age) and 16 postmenopausal (57 ± 4.79 years of age) women took part in the study. The young volunteers selected presented regular menstrual cycles and had not been using anticonceptionals for at least 6 months. The postmenopausal state was defined by the absence of a spontaneous menstrual cycle for at least 1 year, and serum follicular-stimulating hormone levels above 30 mIU/mL. None of the postmenopausal women were using hormone therapy.

All volunteers were in good health based on clinical and physical examination and laboratory tests that included a standard electrocardiogram (ECG), maximum exercise test conducted by physician, total blood count, urinalysis, and clinical biochemical screening tests (serum glucose, uric acid, total cholesterol and fractions, triglycerides and concentration of thyroid-stimulating hormone). None of the volunteers had respiratory, metabolic or cardiovascular diseases. Smokers and users of any kind of medication were excluded from the study. The oxygen uptake data ($\dot{V}O_2$) obtained at peak of the ergospirometric test were used to better characterize the subjects and to obtain the aerobic classification. According to the American Heart Association (21), all volunteers studied had a "very low level" of aerobic capacity, characterizing their sedentary life style.

The volunteers were instructed about the objectives and experimental procedures of the study, its experimental protocol and its non-invasive character. After agreeing to participate, all volunteers gave written informed consent. The Ethics Committee of the University Hospital, Medical School of Ribeirão Preto, University of São Paulo, and the Ethics Committee of the Federal University of São Carlos, SP, Brazil, approved the study.

#### Experimental procedure

The experimental procedures were performed in a climatically controlled laboratory where temperature and relative air humidity were kept at around 23ºC and 60%, respectively. Volunteers familiarized themselves with the laboratory environment and the experimental protocol that they would undergo. On the day of the test they were questioned concerning their health condition, if they had had a good night's sleep and if they had followed the instructions given on the day before, such as no ingestion of alcoholic drinks or of stimulants (coffee, tea, soft drinks), and had done no extenuating physical activity.

The experimental protocol was always applied at the same time of day (morning) taking into consideration the circadian influences on the responses of the variables studied. The volunteers were studied during the follicular phase of the menstrual cycle in order to avoid the influence of hormonal fluctuation on the cardiorespiratory variables measured.

Before the measurements the volunteers rested for 15 min in the supine position, and their arterial blood pressure and HR were measured to determine whether their basal condition was satisfactory for the experiment.

#### Experimental protocol

The experiment consisted of a continuous physical exercise test of the ramp type (CT-R), performed on a cycloergometer with electromagnetic braking (Quinton Corival 400, Seattle, WA, USA), at a bench height regulated to permit knee flexing of 5 to 10 degrees. The volunteers were instructed not to perform an isometric contraction while holding onto the handlebar of the bicycle and to maintain the pedaling rate at 60 rpm.

The CT-R consisted of 1 min pre-testing in the sitting resting position on the cycloergometer followed by a 4-min warm-up period at 4 W and workload increases of 10 to 20 W/min, until physical exhaustion. Workload increases were determined for each volunteer according to the formula proposed by Wasserman et al. (2).

During the test, the volunteers were monitored on the CM5 lead. ECG and HR were obtained beat-to-beat from a one-channel heart monitor (TC 500, ECAFIX, São Paulo, SP, Brazil) and processed with an analog-to-digital converter Lab PC + (National Instruments, Co., Austin, TX, USA), which acts as an interface between the heart monitor and a microcomputer (Pentium III 500 MHz). The ECG signal was recorded in real time after analog-to-digital conversion at a sampling rate of 500 Hz, and the R-R intervals (ms) and HR (bpm) were calculated on a beat-to-beat basis using specific software (22).

Ventilatory and metabolic variables, such as $\dot{V}O_2$, carbon dioxide output ($\dot{V}CO_2$) and

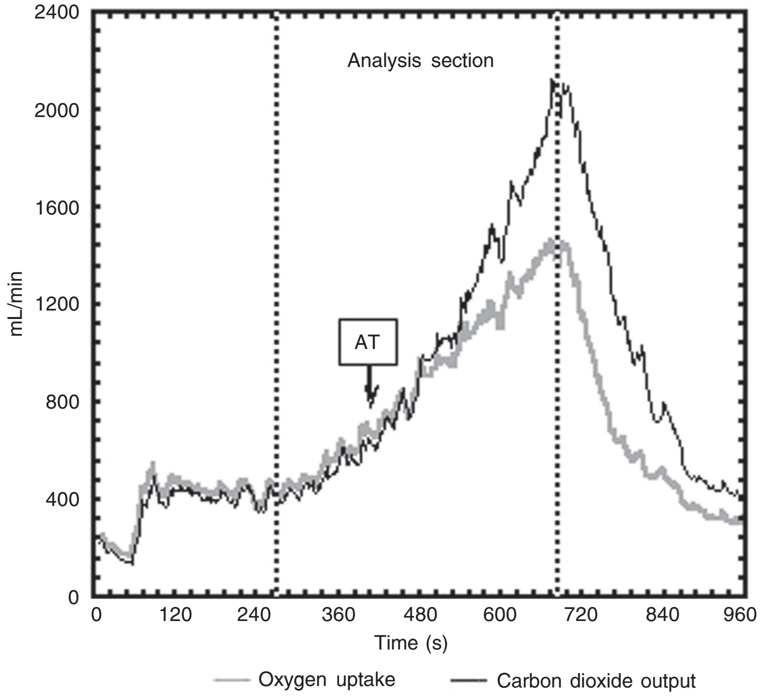


Figure 1. Oxygen uptake and carbon dioxide output responses during the continuous physical exercise dynamic test of the ramp type by one of the volunteers studied. The arrow indicates the time (s) of the disproportionate increase in carbon dioxide output relative to oxygen uptake in the anaerobic threshold (AT) determination.

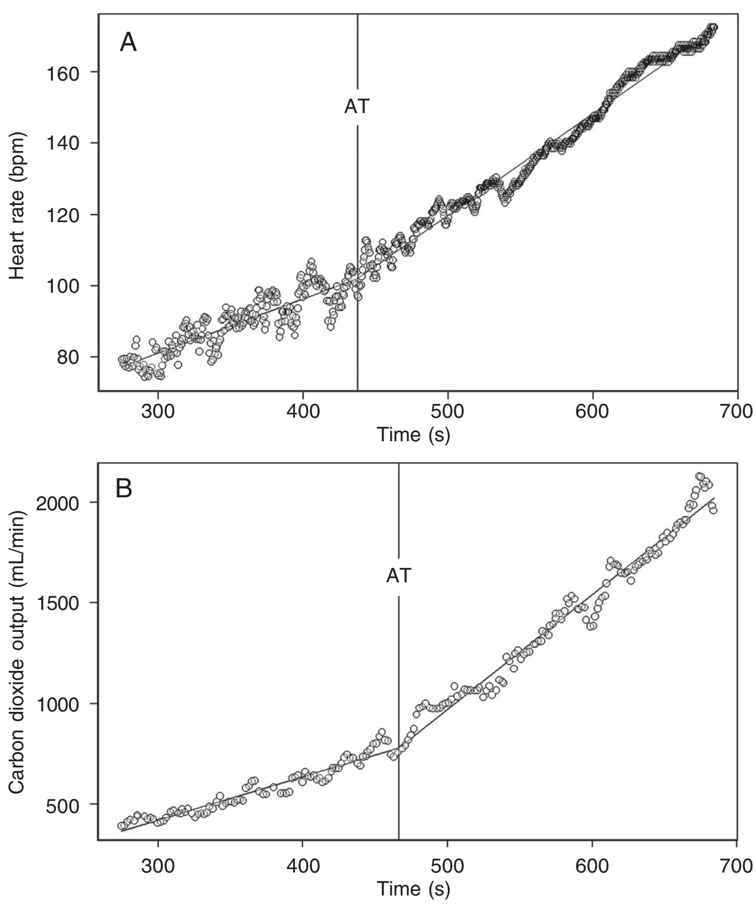


Figure 2. Illustration of the break point determined by the mathematical linear bi-segmental model applied to heart rate (A) and carbon dioxide output (B) data, obtained during a continuous physical exercise test of the ramp type by one volunteer whose data are presented in Figure 1. AT = anaerobic threshold.

minute ventilation ($\dot{V}E$), were obtained breath-to-breath during the CT-R by means of a system measuring expired gases (CPX/D, Medical Graphics, St. Paul, MN, USA) which was calibrated before each test. These variables were subsequently processed and calculated as moving means after every eight respiratory cycles for better kinetic observation of the responses during the exercise.

### Anaerobic threshold determination

Two methods were employed for AT determination at CT-R: 1) graphic visual analysis of the responses of ventilatory and metabolic variables was performed by three observers with proven experience in the application of the procedures used for this purpose. The criterion for AT quantification was graphic observation at the time of a disproportionate $\dot{V}CO_2$ increase in relation to the $\dot{V}O_2$ linear increase on the ergospirometer monitor (Figure 1). This method was based on the V-slope method described by Wasserman and McIlroy (5) which is considered to be a gold standard method. The AT value was considered as the mean of the data obtained from the analysis by three observers. 2) The mathematical model of bi-segmental linear regression was implemented using Hinkley's algorithm applied to HR (MMH-HR) and $\dot{V}CO_2$ (MMH-$\dot{V}CO_2$) data. This permits analysis of the $\dot{V}CO_2$ and HR data series by the maximum similarity method that identifies the moment when a change occurs in the response pattern of $\dot{V}CO_2$ and HR data collected during CT-R performance, and detects the intersection point of two phases adjustment of data sets (Figure 2).

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 respiratory compensation point (RCP) or up to the end of exercise when the volunteer did not present the RCP.

### Statistical analysis

The power output (W), HR (bpm), $\dot{V}O_2$ (mL kg$^{-1}$ min$^{-1}$), $\dot{V}O_2$ (mL/min), $\dot{V}CO_2$ (mL/min), and $\dot{V}E$ (L/min) at AT determined by the two methods used did not present normal distribution upon application of the Kolmogorov-Smirnov test. Therefore, the non-parametric Friedman repeated measurements, Dunn's *post-hoc* and Spearman correlation tests were used, with the level of significance set at 5%.

## Results

No significant differences in weight, height and HR at rest were observed between groups. The subjects of the young group had lower body mass index, blood pressure and a higher $\dot{V}O_2$ peak than middle-aged group ($P < 0.05$; Table 1).

Power output, HR, $\dot{V}O_2$, $\dot{V}CO_2$, and $\dot{V}E$ data, at the AT level, did not differ significantly ($P > 0.05$) between the graphic visual, MMH-HR and MMH-$\dot{V}CO_2$ methods, for both young and postmenopausal groups (Table 2).

Concerning the two methods used to determine AT, a significant correlation of $\dot{V}O_2$, in mL kg$^{-1}$ min$^{-1}$, was detected between the gold

Table 1. Age and anthropometric and clinical characteristics of the subjects studied.

| | Young group (N = 13) | Postmenopausal group (N = 16) |
|---|---|---|
| Age (years) | 24 ± 2.63 | 57 ± 4.79* |
| Weight (kg) | 57 ± 6.02 | 62 ± 9.91 |
| Height (cm) | 163 ± 6.0 | 157 ± 5.0 |
| Body mass index (kg/m$^2$) | 21.61 ± 2.55 | 25.10 ± 3.67* |
| Systolic blood pressure (mmHg) | 99 ± 8.62 | 121 ± 16.92* |
| Diastolic blood pressure (mmHg) | 67 ± 5.55 | 77 ± 9.81* |
| Heart rate (bpm) | 72 ± 8.64 | 67 ± 5.75 |
| $\dot{V}O_2$ peak (mL kg$^{-1}$ min$^{-1}$) | 22.67 ± 3.11 | 14.33 ± 2.73* |
| Follicle-stimulating hormone (mIU/mL) | 3.97 ± 2.18 | 70.6 ± 50.97* |
| Serum estradiol level (pg/mL) | 80.0 ± 51.87 | 23.4 ± 5.94* |
| Menstrual cycle (days) | 29 ± 1.5 | - |
| Years of menopause | - | 9 ± 5.48 |

Data are reported as means ± SD. $\dot{V}O_2$ = oxygen uptake.
*P < 0.05 compared to the young group (unpaired *t*-test).

standard method and the MMH applied to HR ($r_s$ = 0.75) and $\dot{V}CO_2$ ($r_s$ = 0.78) data for the subjects as a whole (N = 29; Figure 3).

## Discussion

Changes in cardiorespiratory response pattern during dynamic exercise are mediated by muscle metabolic activity (2). Hinkley's mathematical algorithm of the bi-segmental linear regression model (20) was used to identify the moment at which these changes occur.

The principle of the proposed mathematical model is based on the fitting of two straight lines by maximum similarity of data sets analyzed. The first straight line adjusted by the algorithm represents the variable behavior at the beginning of physical effort up to the AT. The second straight line corresponds to the change in variable response pattern, which is characterized by an increase in its rate of variation until RCP or physical exhaustion. Thus, the break point of the two straight lines was considered to be the moment when AT occurs (Figure 2).

The mathematical model used in the present investigation allowed the determination of the break point in the dynamic behavior pattern of $\dot{V}CO_2$ and HR data that occurs during physical exercise, as well as its interactions with the level of physical effort.

The aging physiologic process is marked with several changes that influence individual aerobic capacity (23,24), as well as the magnitude of the cardiorespiratory response to exercise. Due to this process, our objective was to evaluate whether these alterations could affect the accuracy of the proposed mathematical model for women in any age group.

Table 2. Cardiorespiratory parameters at the anaerobic threshold determined by a graphic visual method (VM) and by the mathematical model of Hinkley's bi-segmental linear regression applied to heart rate (MMH-HR) and carbon dioxide output (MMH-$\dot{V}CO_2$) data for the young and postmenopausal groups.

| Group | VM | MMH-HR | MMH-$\dot{V}CO_2$ |
|---|---|---|---|
| Young (N = 13) | | | |
| Power output (W) | 51 | 41 | 44 |
| Heart rate (bpm) | 115 | 110 | 113 |
| $\dot{V}O_2$ (mL $kg^{-1}$ $min^{-1}$) | 11.76 | 10.46 | 11.18 |
| $\dot{V}O_2$ (mL/min) | 661 | 591 | 628 |
| $\dot{V}CO_2$ (mL/min) | 640 | 550 | 576 |
| $\dot{V}E$ (L/min) | 18.9 | 16.8 | 17.8 |
| Postmenopausal (N = 16) | | | |
| Power output (W) | 33 | 37 | 37 |
| Heart rate (bpm) | 103 | 108 | 104 |
| $\dot{V}O_2$ (mL $kg^{-1}$ $min^{-1}$) | 9.10 | 9.36 | 9.29 |
| $\dot{V}O_2$ (mL/min) | 584 | 628 | 605 |
| $\dot{V}CO_2$ (mL/min) | 579 | 617 | 581 |
| $\dot{V}E$ (L/min) | 18.3 | 20.6 | 19.4 |

Data are reported as medians. $\dot{V}O_2$ = oxygen uptake; $\dot{V}CO_2$ = carbon dioxide output; $\dot{V}E$ = minute ventilation.
P > 0.05 (Friedman repeated measurements and *post hoc* Dunn test).

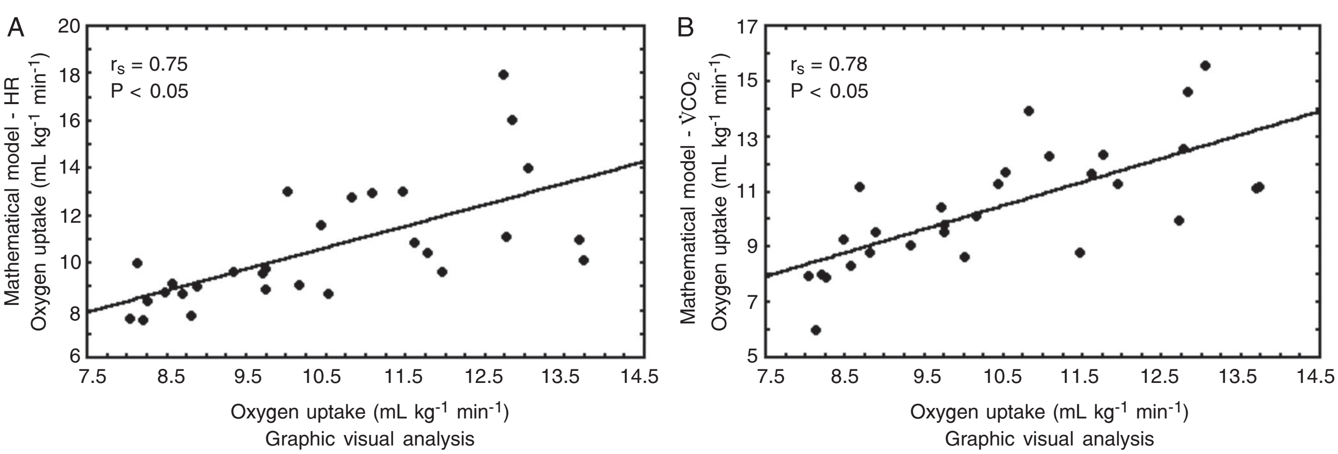


Figure 3. Correlation of the oxygen uptake data obtained by the graphic visual method with the mathematical model applied to heart rate (HR, in A) and carbon dioxide output ($\dot{V}CO_2$, in B) for the subjects as a whole (Spearman correlation test).

The comparison analysis of AT values determined by visual and mathematical methods showed a good fit in detection of the break point in response pattern of $\dot{V}CO_2$ and HR in relation to time, for both young and postmenopausal groups, because the power output, HR, $\dot{V}O_2$, $\dot{V}CO_2$, and $\dot{V}E$ values obtained at AT determined by visual method were similar to the mathematical model.

For analysis of the correlation between the methods used, the two groups of volunteers were studied as a whole. The analysis of $\dot{V}O_2$ data (mL $kg^{-1}$ $min^{-1}$) showed a statistically significant correlation between the gold standard method and the bi-segmental linear regression mathematical model when applied to the HR ($r_s = 0.75$) and $\dot{V}CO_2$ ($r_s = 0.78$) data.

Thus, our findings showed that the mathematical model was adequate for AT determination in sedentary healthy women. Other studies from our laboratory found that the ventilatory AT (gold standard method) was very close to the AT determined by the autoregressive integrated moving average model applied to middle-aged healthy men (10).

Studies that compare the analysis of ventilation and metabolic variables by trying to determine the index of these variables that best corresponds to AT estimated by the blood lactate method are available in the literature (6,7,16,25-27). Other studies have identified AT using non-invasive methods by analysis of cardiorespiratory variables, developing mathematical models applied to $\dot{V}CO_2$, such as multi-segmental linear regression (18) and linear-linear and linear-quadratic bi-segmental regressions (8).

Conconi et al. (3) presented a pioneering proposal to verify the relationship between running speed and loss of HR linearity in runners for AT determination. Nevertheless, this method was not able to determine the AT, but it permitted the quantification of RCP. After that, other investigators conducted research based on Conconi's study in order to improve the analytical methodologies, using the third-order curvilinear regression method (19) and mathematical model of linear adjustment of HR data (12,13).

The mathematical model used in the present study showed to be a promising tool for AT determination, since it constitutes a non-invasive, objective and semi-automatic method. Furthermore, it decreases the variations of human acuity evoked by graphic visual analysis, optimizing the process for the determination of this physiological parameter.

The results of this study showed that the mathematical linear regression bi-segmental model can be used for AT identification based on $\dot{V}CO_2$ data as well as on HR response. This method permits AT determination in clinics and consulting rooms by analysis of HR, which is a simple and easily obtained variable, permitting an optimization of this process.

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