Comparison between anaerobic threshold determined by ventilatory variables and blood lactate response in cyclists

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ABSTRACT
Many investigations have shown that the coincidence between the ventilatory thresholds and those thresholds using the lactate response does not happen all of the time, suggesting that there is no relationship between the cause-effect between these phenomena. Thus, the present study had as main purpose to compare and correlate the Oxygen consumption (VO2), the power (W), and the heart rate (HR) values attained using protocols to determine the Ventilatory Threshold (VT) and the Individual Anaerobic Threshold (IAT). The sampling was constituted by eight State and National level cyclists (age: 27.88 ± 8.77 years; body mass: 65.19 ± 4.40 kg; height: 169.31 ± 5.77 cm). The IAT was determined starting from a three minutes 50 W warm up with progressive increases of 50 W.min-1 up to achieving the voluntary exhaustion, when the blood was collected in the last 20 seconds of each phase, and during the recovering period. In order to determine the VT, it was used the same protocol used to determine the IAT, but without performing the blood collection. The VT was identified through the changes in the pulmonary ventilation, as well as of the ventilatory equivalent of the O2 and CO2. The t-Student test showed no significant statistical difference in any of the attained variables. The associations found were high and significant. The VO2 (ml.kg-1.min-1), P (W), and HR (bpm) corresponding to the VT and IAT, as well as the associations between variables were respectively: 48.00 ± 3.82 vs. 48.08 ± 3.71 (r = 0.90); 256.25 ± 32.04 vs. 246.88 ± 33.91 (r = 0.84); 173.75 ± 9.18 vs. 171.25 ± 12.02 (r = 0.97). According to the results attained, it can be concluded that the IAT and the VT produce similar ventilatory thresholds, and this has originated the term “aerobic-anaerobic transition” introduced by Kindermann et al.(2). The first transition point is identified as Aerobic Threshold (AeT) that reflects the exercise intensity corresponding to the maximal steady state of the blood lactate (MSSL). The authors suggest that the first transition corresponds to the AnT proposed by Wasserman and McIlroy, or the Ventilatory Threshold (VT) proposed by Kindermann et al.(2). The second transition point is considered the breathing compensatory point of the MSSL, or the Ventilatory Threshold 2 (VT2). These different terminologies for correlate phenomena have been caused some confusion in the area of the exercise physiology. In order to determine the intensities corresponding to the AeT and the AnT, Kindermann et al.(2) adopted fixed concentrations of 2 and 4 mmol.1-1 of the blood lactate, respectively, in an incremental exercise protocol. The majority of the researchers use fixed 4 mmol.1-1 blood lactate concentrations to determine the MSSL, and they have proposed several terminologies to identify such phenomenon. Heck et al.(10) justify the option for that fixed blood lactate concentration (4 mmol.1-1) as the majority of individuals presents such exercise intensity, the maximal ability to remove the produced lactate. Nevertheless, in that same study, it was verified that the MSSL can occur in blood lactate concentrations within a 3.1 and 5.54 mmol.1-1 range. Similar results have been verified by Stegmann et al.(16), who found different individual values of the blood lactate upon the identification of the MSSL in an incremental test in which they varied between 1.4 and 7.5 mmol.1-1. Having in mind the high inter-individual variability found in the results, the authors introduced the term Individual Anaerobic Threshold (IAT), which is an identifying method for the MSSL that does not respect the fixed lactate concentration, and it may be employed on running, cycle ergometer, rowgometer, as well as for performance assessment, training prescription and control.(3,4,7,9).

INTRODUCTION
In the last decades, the metabolic thresholds have been the target of several investigations within the exercise physiology, and they are considered extremely relevant parameters, more important than the maximal oxygen consumption to prescribe the training intensity(1-3), to control the effects of the training(4,5), and to predict the physical performance(6-8).

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The term Anaerobic Threshold (AnT) was introduced by Wasserman and McIlroy(9), and it is defined as the strength intensity anterior to the exponential increase factor of the blood lactate compared to the resting levels. Later, it was verified that there are two thresholds, and this has originated the term “aerobic-anaerobic transition” introduced by Kindermann et al.(2). The first transition point is identified as Aerobic Threshold (AeT) that reflects the exercise intensity corresponding to the maximal steady state of the blood lactate (MSSL). The authors suggest that the fist transition corresponds to the AnT proposed by Wasserman and McIlroy, or the Ventilatory Threshold (VT) introduced by Kindermann et al.(2). The second transition point is considered the breathing compensatory point of the MSSL, or the Ventilatory Threshold 2 (VT2). These different terminologies for correlate phenomena have been caused some confusion in the area of the exercise physiology. In order to determine the intensities corresponding to the AeT and the AnT, Kindermann et al.(2) adopted fixed concentrations of 2 and 4 mmol.1-1 of the blood lactate, respectively, in an incremental exercise protocol. The majority of the researchers use fixed 4 mmol.1-1 blood lactate concentrations to determine the MSSL, and they have proposed several terminologies to identify such phenomenon. Heck et al.(10) justify the option for that fixed blood lactate concentration (4 mmol.1-1) as the majority of individuals presents such exercise intensity, the maximal ability to remove the produced lactate. Nevertheless, in that same study, it was verified that the MSSL can occur in blood lactate concentrations within a 3.1 and 5.54 mmol.1-1 range. Similar results have been verified by Stegmann et al.(16), who found different individual values of the blood lactate upon the identification of the MSSL in an incremental test in which they varied between 1.4 and 7.5 mmol.1-1. Having in mind the high inter-individual variability found in the results, the authors introduced the term Individual Anaerobic Threshold (IAT), which is an identifying method for the MSSL that does not respect the fixed lactate concentration, and it may be employed on running, cycle ergometer, rowgometer, as well as for performance assessment, training prescription and control.(3,4,7,9).

Several researchers have investigated the relationship between the IAT and other protocols aiming to identify the MSSL, but in those studies, the blood lactate response was determined using the direct method. The determination of the blood lactate response by means of the direct method requires a scheduled blood collection, thus, it is necessary to use non-invasive methods to identify that phenomenon. Some studies involving non-invasive methods to determine the IAT have been developed. Nevertheless, it was found no coincidence in the exercise intensity attained by the IAT compared to the critical power(19) and the deflection point of the heart rate proposed by Conconi(27).
The non-invasive method that allows identify the MSSL during the incremental load exercise, involves the VT estimate. Nevertheless, the VT and the thresholds coincidence using lactate does not occur all the time, suggesting that there is no cause-effect relationship between these phenomena. The findings from studies that have analyzed the relationship between the IAT and the VT are quite controversial\(^{31,28,29}\).

Based on these facts, the purposes of the present study were: to set comparisons between the Oxygen consumption (\(\text{VO}_2\)), the intensity (\(W\)), and the heart rate (HR) values during the accomplishment of the protocols to determine the IAT and the VT in cycling athletes, and later, to verify the possible associations between parameters attained by both methods.

METHODS

Subjects

It participated in this study eight male cycling athletes at the state and national levels competing in the Bike Speed (\(n = 4\)) and Mountain Bike (\(n = 4\)) categories. The general characteristics of the sampling are presented on table 1. As pre-requirements to be admitted in the trial, athletes should have at least two years experience in regional or state competitions. After being examined by a physician, every individual received information on the goals of the study and the procedures which they would be submitted to, and they signed a free clarified consent. The study was developed at the CeMENutri (Center of Metabolism in Exercise and Nutrition), and it was approved by the Ethics Committee in Research of the Botucatu School of Medicine/UNESP, SP.

Dietetic control

Aiming to avoid the trial would suffer any kind of interference in the test results as to the energetic substrate availability\(^{30,31}\), athletes had a nutritional follow-up along the whole study.

From the application of a food questionnaire (recalling 24 hours of their food habits), the nutritionists team elaborated ordinary diet schedules as to the food habits of the assessed individuals. They were instructed to follow such diet along the whole period of the trial. Furthermore, it was elaborated a standard breakfast to be consumed two hours before the test accomplishment. Also, all individuals were instructed to avoid the intake of caffeinated products 24 hours before the tests, as those substances could influence the results\(^{32}\). The information on the quantity and quality of the consumed foods was processed by means of the Virtual Nutri version 1.0 nutritional analysis software.

Experimental outlining

In the first step of the trial, individuals came to the laboratory to have their medical examination and anthropometric measurements, in order to characterize the sampling, and to have an interview with the nutritionists, when they received the food intake guidelines to be followed along the whole period of the trial. Furthermore, it was scheduled a standard breakfast to be consumed two hours before the test accomplishment. Also, all individuals were instructed to avoid the intake of caffeinated products 24 hours before the tests, as those substances could influence the results\(^{32}\). The information on the quantity and quality of the consumed foods was processed by means of the Virtual Nutri version 1.0 nutritional analysis software.

From the second step of the trial on, the individuals came to the laboratory in predefined hours, when they were submitted to the test to determine the IAT and the VT\(_2\), which were randomly performed. All the tests (pre-trial, IAT, and VT\(_2\)) were applied with a 72 hours interval between them. Individuals were instructed not to perform their physical activities 24 hours prior to the accomplishment of each test, in order to avoid any interference.

Ergospirometry

The incremental tests were performed in an electromagnetic ergospirometric cycle (Corival 400, Quinton\(^{\text{®}}\), Bothell, USA). The ventilatory variables were continuously measured in an open circuit ergo-spirometric system (QMC\(^{\text{TM}}\) 90 Metabolic Cart, Quinton\(^{\text{®}}\), Bothell, USA) using the breath-by-breath technique. At the beginning of each test, the gauging was performed through a Hans Rudolf 5530 3-liters calibration syringe and a mix of 26% \(\text{O}_2\) gas with a \(\text{N}_2\) and 4% \(\text{CO}_2\) and 16% \(\text{O}_2\) balance (White Martins Praxair, Inc. São Paulo, Brazil).

The analysis was processed on an IBM computer through the calculations of the minute ventilation (VE), the \(\text{VO}_2\), the carbon dioxide production (\(\text{VCO}_2\)), and the relationship between the carbon dioxide production and the oxygen consumption (R).

The HR was measured through a cardiofrequencymeter (Vantage NV, Polar Electro OY, Finland) with an every 5 seconds record, uploading on a software (Polar Precision Performance\(^{\text{TM}}\), Finland) for later analysis. The HR corresponding to different loads was determined from the recorded values of the last five seconds of each phase. The variables of the environmental temperature and the relative air humidity were kept between 21 and 24°C, and 40 and 60%, respectively.

IAT

To determine the IAT, initially, the individuals performed a three minute warm up at a 50 W load, and next, the incremental test started with a 50 W increase to the load every three minutes, keeping the 70 revolution per minute cadency. During the incremental test, the individuals were verbally encouraged to go on up to the voluntary exhaustion. The blood collection was performed in the ear lobe on a resting condition (pre-strength) in the final 20 seconds of each load up to the exhaustion, and at three, five and ten minutes after the test finished.

From the construction of a graphic representing the blood lactate values in each phase of the incremental test and during the passive recovering (figure 1), the IAT was determined following the procedures proposed by Stegmann et al.\(^{16}\).

\[ \text{Fig. 1} - \text{Determining the intensity corresponding to the IAT} \]

\[ \text{Fig. 2} - \text{Identification of the VT and VT}_2 \text{ according to the VE/VO}_2 \text{ and VE/VCO}_2 \]
The VT was identified through the application of the same protocol used to determine the IAT, but in this situation, the blood collection was not performed. The VT₂, or breathing compensatory point, had a double identification through the use of the ventilatory equivalent of the Oxygen (VE/VO₂), the ventilatory equivalent of the carbon dioxide (VE/VCO₂) considering the sudden increase in the VE/VCO₂ according to the criteria proposed by McLellan (11).

Figure 2 illustrates the identification of the thresholds; nevertheless, it was performed in this study only the VT₂.

Biochemical analysis

It was collected 25 µl of the ear lobe blood through a previously gauged heparinized glass capillary, and they were immediately transferred to a 1.5 ml “Ependorf” type polyethylene microtubes containing 50 µl of a 1% sodium fluoride solution. Next, the samples were stored at −70°C. The lactate analysis was performed using an electroenzimatic analyzer (YSL 2300 STAT Yellow Spring Co., USA), and the values were expressed in mmol.l⁻¹.

Statistical treatment

The results were gathered according to the mean values and standard deviation using the statistical package Statistica 6.0® (STATSOFT INC., USA). Upon the application of the Shapiro Wilk test, it was verified that the data distribution were normal. Thus, the variables attained in the protocols contrasted from the t-Studen test for dependent sampling. The linear Pearson correlation coefficient was employed to verify the associations between the variables attained in the protocols, in order to determine the IAT and the VT₂. The significance level adopted for every analysis was 1%.

RESULTS

Table 1 presents the general features of the sampling.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Mean</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>27.88</td>
<td>8.97</td>
</tr>
<tr>
<td>Body mass (kg)</td>
<td>65.19</td>
<td>4.40</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>169.31</td>
<td>5.77</td>
</tr>
<tr>
<td>Time of practice (years)</td>
<td>6.17</td>
<td>4.7</td>
</tr>
<tr>
<td>Weekly training volume (km)</td>
<td>146.7</td>
<td>34.5</td>
</tr>
<tr>
<td>Weekly training frequency (days)</td>
<td>5.00</td>
<td>0</td>
</tr>
</tbody>
</table>

Table 2 presents the values for the VO₂max, the intensity at the moment of the exhaustion (WVO₂max), and the FCmax attained in both protocols used in this study (IAT and VT₂). It was found no significant differences compared to the above described variables at the exhaustion moment for the IAT and VT₂ tests. On that same table, it can be observed the blood lactate values at the intensity corresponding to the IAT and the maximal load.

The results of the VO₂, W and HR corresponding to the VT₂ and the IAT are presented on table 3. The t-Student test did not identify significant differences between the VO₂, W and HR values attained in the VT₂ and IAT protocols.

<table>
<thead>
<tr>
<th>Variables</th>
<th>VT</th>
<th>IAT</th>
</tr>
</thead>
<tbody>
<tr>
<td>VO₂Threshold (ml.kg⁻¹.min⁻¹)</td>
<td>48.00 ± 1.35</td>
<td>48.08 ± 1.31</td>
</tr>
<tr>
<td>WVO₂max (W)</td>
<td>256.25 ± 11.32</td>
<td>246.88 ± 11.98</td>
</tr>
<tr>
<td>HRThreshold (bpm)</td>
<td>173.75 ± 3.2</td>
<td>171.25 ± 4.25</td>
</tr>
</tbody>
</table>

VT = ventilatory threshold; IAT = individual anaerobic threshold; VO₂Threshold = Oxygen consumption at the thresholds; WVO₂Threshold = Intensity corresponding to the thresholds; HRThreshold = heart rate corresponding to the thresholds.

* Significant difference (p < 0.01).

The linear regression between the VO₂, the HR and W attained in both methods are presented in figures 3, 4 and 5, respectively. The associations were high and significant in every analyzed variable.

**Fig. 3** - Linear regression between the VO₂ at the individual anaerobic threshold (IAT) and the ventilatory threshold 2 (VT)

**Fig. 4** - Linear regression between the heart rate (HR) at the individual anaerobic threshold (IAT) and the ventilatory threshold 2 (VT)
DISCUSSION

The IAT is defined as the higher metabolic rate where the blood lactate concentration is kept in a balanced state during prolonged exercises, and the elimination of the blood lactate is maximal, being equivalent to the diffusion rate of the muscular compartment to the blood\(^{[16]}\). Therefore, it might be considered that the IAT protocol is capable to determine the intensity corresponding to the MSSL.

In such sense, several studies have found the MSSL during the long endurance rectangular test at the intensity corresponding to the IAT, making such relationship evident\(^{[4,20,23,26,33]}\). The IAT is defined as the higher metabolic rate where the blood lactate concentration is kept in a balanced state during prolonged exercises, and the elimination of the blood lactate is maximal, being equivalent to the diffusion rate of the muscular compartment to the blood\(^{[16]}\). Therefore, it might be considered that the IAT protocol is capable to determine the intensity corresponding to the MSSL.

Initially, the criteria adopted to identify the ventilatory thresholds were the breaking points of the VE related to the VO\(_2\). Later, besides the previously mentioned criterion, it was suggested the use of other variables, such as VE/VO\(_2\), VE/CO\(_2\), and R\(^{[40,41]}\). Bascially, the aim was to identify an incremental load protocol when there is an increase in the VE/VO\(_2\), and the O\(_2\) pressure with no alterations on the VE/CO\(_2\), and on the CO\(_2\) pressure. To some authors, such intensity corresponds to the VT\(^{[11]}\). From this point on, the increase in the exercise intensity will cause a metabolic acidosis, resulting in a decrease in the pH, and consequently an increase on the VE/CO\(_2\) and on the CO\(_2\). That second point is considered the VT\(^{[11]}\), or the respiratory compensatory point.

Related to the individual’s physical condition, the mechanisms involved in the occurrence of the AnT seem to be the same both in athletes and in non-trained individuals. However, the point where the phenomenon occurs is different among them (Wyatt, 1999).

Another important aspect that cannot be disregarded is the protocol adopted to determine the ventilatory thresholds. The optimum protocol is the one that allows the researcher to observe the inflexion point of the EV/VO\(_2\) and the region of the isocapnic tamponage (increase in the EV/VO\(_2\), with no modifications in the EV/ CO\(_2\)). Thus, Davis\(^{[43]}\) suggests the use of incremental protocols constituted by one minute endurance phases. In this study, the protocol employed to determine the VT was similar to that adopted to identify the IAT, that means, three minutes endurance each phase. According to McLellan\(^{[11]}\), that second threshold seems not to suffer any influence on the endurance of the phases (1, 3, or 5 minutes).

As it can be seen, there is a great amount of criteria and terminologies used to identify the metabolic thresholds. Thus, it is fundamental to make judicious observation of the protocol adopted to determine the blood lactate response, mainly aiming to prescribe the training intensity.

It is important to point out that one of the limitations of this study is related to the reduced sampling. In such sense, Stone et al.\(^{[44]}\) recently pointed out that it is required to make a distinction between the concepts of Exercise Science and Sports Science. Generally, the literature has several publications related to the Exercise Science. Nevertheless, there is a scarcity of publications related to the sports itself, that means, the Sport Science, understood as the one developed with the purpose to propitiate the sportive performance increment through the application of methods and scientific principles to the training assessment, controlling, and prescription\(^{[44]}\).
CONCLUSION

Based on the results found in this study, it can be concluded that the protocols to determine the IAT and VT give similar VO₂, intensity and HR values, even presenting high correlations between these variables, favoring the adoption of the VT as this is a non-invasive method to determine the anaerobic threshold in cyclists.

All the authors declared there is not any potential conflict of interests regarding this article.

REFERENCES