

## Research article

# EFFECT OF TIME OF DAY ON THE RELATIONSHIP BETWEEN LACTATE AND VENTILATORY THRESHOLDS: A BRIEF REPORT

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### ABSTRACT

The purpose of this investigation was to study the effect of time of day on the relationship between lactate (LT) and ventilatory thresholds (VT) of pulmonary oxygen uptake ( $VO_2$ ). Seven moderately active male volunteers (26.3±3.0 years, 1.74±0.08 m, 76±5 kg) performed a maximal incremental test (increases of 30 W every 2 min) on a cycle ergometer on consecutive days at 0900 h, 1400h and 1900 h in a randomized fashion. The anaerobic threshold was determined using both ventilatory gas analysis and blood lactate measures. Each of the following variables was recorded both at VT and the LT; heart rate (HR, beats.min<sup>-1</sup>), minute ventilation (VE, L.min<sup>-1</sup>), respiratory exchange ratio (RER), time to threshold (Time, sec), oxygen uptake ( $VO_2$ , ml.kg<sup>-1</sup>.min<sup>-1</sup>) and  $VO_2$  as a percentage of maximal oxygen uptake (% $VO_{2max}$ ). The correlations between VT and LT variables analyzed by Pearson product moment correlations for each time of day. ANOVA was used to compare the data obtained at different times of the day. There were no significant differences for the data related to time of day either for ventilatory gas analysis or lactate measurements. The correlation coefficients between VT and LT variables were moderate to high ( $r=0.56-0.94$ ) for time of day. However, the correlations for HR,  $VO_2$ , and % $VO_{2max}$  ( $r=0.81-0.94$ ) were slightly stronger compared with Time, VE and RER ( $r=0.56-0.88$ ). It was concluded that, the data at VT and LT were not influenced by time of day.

**KEY WORDS :** Anaerobic threshold, circadian rhythm.

### LAKTAT VE SOLUNUMSAL EŞİK ARASINDAKİ İLİŞKİ ÜZERİNE GÜNÜN ZAMANININ ETKİSİ: KISA RAPOR

#### ÖZET

Bu araştırma ile günün farklı zamanının laktat (LT) ve solunumsal eşik (VT) arasındaki ilişki üzerine olan etkisini incelemek amaçlandı. Yedi orta düzeyde aktif erkek gönüllü (26.3±3.0 yıl, 1.74±0.08 m, 76±5 kg) birbirini takip eden günlerde rastgele sıra ile saat 0900, 1400 ve 1900'da bisiklet ergometresinde artan yüklerin (2 dakikada bir 30 W) kullanıldığı maksimal bir test yaptı. Anaerobik eşik solunumsal gaz analizi ve laktik asit ölçümleri ile belirlendi. VT ve LT için takip eden değişkenler kaydedildi; kalp atım sayısı (HR, sayı.dak<sup>-1</sup>), ventilasyon hacmi (VE, L.min<sup>-1</sup>), solunum oranı ( $VCO_2 / VO_2$ , RER), eşığe ulaşılan zaman (Time, san), oksijen alımı ( $VO_2$ , ml.kg<sup>-1</sup>.dak<sup>-1</sup>) ve  $VO_2$  'nin maksimal oksijen tüketimine ( $VO_{2max}$ ) yüzdesel oranı ( $VO_{2max} \%$ ). VT ve LT için kaydedilen değişkenler arasındaki ilişkiler Pearson korelasyon matrisi ile analiz edildi. Günün farklı zamanında elde edilen değerlerin karşılaştırmasında ise ANOVA kullanıldı. ANOVA sonuçlarına göre günün zamanı ile ilişkili olarak ne solunumsal gaz analizi ne de laktik asit ölçümü için anlamlı farklılıklar yoktu. Test edilen zamanlar için VT ve LT değişkenleri arasındaki korelasyon katsayıları orta ile iyi düzey ( $r=0.56-0.94$ ) arasındaydı. Bununla birlikte Time, VE and RER ( $r=0.56-0.88$ ) ile karşılaştırıldığında HR,  $VO_2$ ,  $VO_{2max}\%$  ( $r=0.81-0.94$ ) için korelasyonlar biraz daha kuvvetli idi. VT ve LT ile ilişkili değerlerin genel olarak değerlendirildiğinde günün zamanından etkilenmediği sonucuna varıldı.

**ANAHTAR KELİMELER:** Anaerobik eşik, sirkadiyan ritim.

## INTRODUCTION

The increase in the blood lactate, above certain exercise intensity, has been recognized since early 20<sup>th</sup> century (Douglas, 1927). This lactate increase has also been termed as the anaerobic threshold (AT) to indicate the pulmonary O<sub>2</sub> uptake (VO<sub>2</sub>) level above which aerobic energy production is supplemented by anaerobic mechanisms (Beaver et al., 1986). Thus, the “anaerobic threshold” (AT) is frequently measured in human performance laboratories to define the athletes’ capacity for endurance exercise.

To determine an individual’s anaerobic threshold, in general, blood lactate measurements can be obtained directly and invasively, or respiratory measures may be used to detect a ‘ventilatory threshold’ non-invasively. Wasserman and McIlroy (1964) and Beaver et al. (1986) described the ventilatory threshold (VT) as a non-invasive method for determining the anaerobic threshold. Since that time, investigators have used both methods of blood lactate (lactate threshold, LT) and ventilatory gases (ventilatory threshold, VT) to define the endurance capacity of athletes. However, the relationship between these two methods is still controversial (Loat and Rhodes, 1993). Several researchers have examined the relationship between VT and LT under different conditions such as glycogen depletion (Neary et al., 1985), and differing training status (Poole and Gaesser, 1985; Burke et al., 1994), and it was reported that the relationship might be causal or coincidental i.e. LT and VT may be coincidental at a given work rate, but one does not necessarily cause the other -e.g. McArdle’s patients. (Gaesser and Poole, 1986; Loat and Rhodes, 1993).

Although, the effects of time of day on VO<sub>2</sub> kinetics (Hill, 1996) and lactate responses to maximal exercise (Deschenes et al., 1998) have been reported previously, the effect of time of day on the relationship between the lactate and ventilatory responses is less known. Therefore, some of the contradictions about the relationship between VT and LT observed in previous studies may be related to the time of day selected for these studies. We aimed to investigate the effect of time of day on the relationship between measures made at VT compared with LT.

## METHODS

### *Subjects*

Seven moderately active male subjects [mean (±SD) age 26.3 (3.0) years, height 1.74 (0.08) m, body mass 76 (5) kg, body fat 15.7 (4.3) %, maximal

oxygen uptake 42.2 (4.2) ml.kg<sup>-1</sup>.min<sup>-1</sup>] volunteered for the study. After being informed about the study and test procedures, and any possible risks and discomfort that might ensue, their written informed consent to participate was obtained in accordance with Helsinki Declaration (WMADH, 2000).

### *Experimental design*

For each volunteer, tests were performed at 09:00 hours, 14:00 hours, and 19:00 hours in a randomized fashion. At least 48 hours separated any two tests scheduled for the same time of day and subjects refrained from exercise for a minimum of 24 hours prior to testing. Volunteers consumed only water for at least 3 h prior to arriving in the laboratory and reported normal sleeping habits for the duration of the study. Each subject completed all tests sessions within 7-10 days.

### *Exercise testing and measurements*

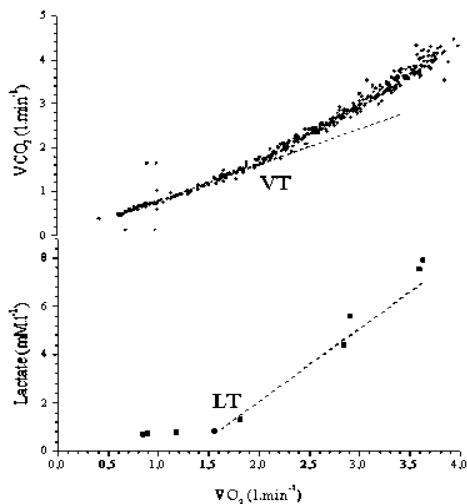
Each subject’s body mass, height and estimated body composition (Womersley and Durnin, 1974) was determined before the first test session. During every test session the subjects arrived in the laboratory 30 min prior to the time set for the cycle ergometer test. Following 20 min supine rest, the subjects’ oral temperature and resting heart rate were measured. Subjects then sat quietly in a chair for 10 min preceding the test and resting blood samples were drawn from a finger to measure resting blood lactate concentration.

All the subjects performed a maximal incremental test on a cycle ergometer following 5 min of warm-up by cycling (Monark 814E, Sweden) at 60 watts (W). The workload during the maximal test was increased 30 W every 2 min from an initial 30 W until volitional subjective exhaustion. The subjects were instructed to maintain pedaling rate as close to 60 rpm as possible and the actual measured pedaling rate was 60±3 rpm for all tests. When the pedaling rate decreased to 55 rpm, subjects were verbally encouraged to maintain the required pedal rate. If the subject could not sustain the target rpm test was terminated. Heart rates were recorded continuously from 4 chest electrodes and monitored via oscilloscope (Cardiovit, Switzerland). Ventilatory parameters were continuously measured “breath by breath” using a metabolic analyzer (SensorMedics 2900C system, USA) during the maximal tests. The criteria for achieving VO<sub>2</sub>max was evaluated as a heart rate within ± 10 beats.min<sup>-1</sup> of the age related maximum (220-age in years), VE/VO<sub>2</sub> values close to 30 Lmin<sup>-1</sup> and respiratory exchange ratio (RER) greater than 1.15. The subjects were also asked to mark relative difficulty level of the exercise at regular intervals according to the Borg scale.

Blood was sampled by finger-stick at rest and during the last 60 seconds of each stage of the maximal test. Blood lactate concentrations were determined using the lactate test strip (Boehringer Mannheim, Germany) and instrument (AccuSport, Boehringer Mannheim, Germany).

#### Lactate and ventilatory threshold determinations

Blood lactate threshold was determined individually for each subject by the method described by Beaver et al. (1985). Ventilatory threshold was determined individually for each subject using V-slope method (Beaver et al., 1986) and other several standard plots as described by Whipp et al. (1986). An experienced observer was asked to confirm the values for both methods. Additionally, in case of disagreement a second independent one was consulted. The results of a representative subject are presented in Figure 1.



**Figure 1.** A representative example of ventilatory threshold (VT) and lactate threshold (LT) determination.

Each of the following variables was recorded at both the VT and the LT; heart rate (HR,  $\text{b}\cdot\text{min}^{-1}$ ), minute ventilation (VE,  $\text{L}\cdot\text{min}^{-1}$ ), respiratory exchange ratio ( $\text{VCO}_2/\text{VO}_2$ , RER), time to threshold (Time, sec), oxygen uptake ( $\text{VO}_2$ ,  $\text{ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ ) and  $\text{VO}_2$  as a percentage of  $\text{VO}_{2\text{max}}$  ( $\%\text{VO}_{2\text{max}}$ ).

#### Statistical analysis

Variables at the LT and VT were compared using a Student's paired  $t$  test. Correlation coefficients between both variables were also calculated for each time of day using a Pearson Product Moment Correlation Matrix. One-way analysis of variance (ANOVA) for repeated measures with Tukey post-hoc tests was used to compare the data obtained at different times of the day. Statistical significance was accepted at  $p < 0.05$ .

## RESULTS

Data obtained during the maximal tests are given in Table 1. These data are values calculated at VT and LT. According to ANOVA results, there were no significant differences between times of the day for these variables either for VT or LT. However, the values for same variables calculated by two methods were significantly different by paired  $t$  test for 0900, 1400 and 1900. The relationships between VT and LT were moderate for exercise time to threshold, RER, VE and  $\text{VO}_2$  %, and were good to high for heart rate and  $\text{VO}_2$  (Table 2). However, the strength of the relationships were not influenced by time of day except VE at 1900 and time to threshold at 1400. Resting oral body temperature at 0900 ( $36.6 \pm 0.2^\circ\text{C}$ ) was significantly lower than the temperature at 1400 ( $36.9 \pm 0.2^\circ\text{C}$ ,  $p < 0.05$ ) and at 1900 ( $37.0 \pm 0.3^\circ\text{C}$ ,  $p < 0.01$ ).

**Table 1.** The results of the variables related to ventilatory threshold (VT) and lactate threshold (LT) at different times of day. Values are mean (SD).

Variables	09:00 hours		14:00 hours		19:00 hours	
	VT	LT	VT	LT	VT	LT
Time (sec)	717 (79)	448 (121) <sup>b</sup>	660 (78)	410 (130) <sup>b</sup>	618 (100)	404 (85) <sup>b</sup>
HR ( $\text{b}\cdot\text{min}^{-1}$ )	166 (14)	144 (17) <sup>b</sup>	159 (13)	141 (15) <sup>b</sup>	152 (16)	137 (13) <sup>b</sup>
VE ( $\text{L}\cdot\text{min}^{-1}$ )	62 (7)	48 (9) <sup>b</sup>	58 (8)	46 (6) <sup>b</sup>	58 (6)	48 (6) <sup>b</sup>
RER	1.07 (0.02)	1.02 (0.02) <sup>b</sup>	1.07 (0.03)	1.01 (0.05) <sup>a</sup>	1.07 (0.04)	1.01 (0.02) <sup>a</sup>
$\text{VO}_2$ ( $\text{ml}\cdot\text{min}^{-1}$ )	2186 (358)	1792 (417) <sup>b</sup>	2066 (301)	1671 (277) <sup>b</sup>	2005 (248)	1681 (181) <sup>b</sup>
$\%\text{VO}_{2\text{max}}$	68 (6)	56 (10) <sup>b</sup>	64 (5)	52 (8) <sup>b</sup>	63 (6)	52 (6) <sup>b</sup>

<sup>a</sup> and <sup>b</sup> denote  $p < 0.01$  and  $0.001$ , respectively, by paired  $t$  test between VT and LT.

No significant differences between the times of day by ANOVA.

HR= heart rate, VE= minute ventilation, RER= respiratory exchange ratio,  $\%\text{VO}_{2\text{max}}$ =  $\text{VO}_2$  relative to  $\text{VO}_2$  max.

## DISCUSSION

The data demonstrated a significant time of day effect on pre-exercise oral temperature, which increased progressively at each of the three time periods during the day. These results were not surprising since gradual increases in resting temperature during awaken hours have been reported consistently (Reilly and Brooks, 1990; Reilly, 1990). The data also demonstrate that the selected variables at LT and VT were not influenced by time of day determined either by ventilatory gas analysis or lactate measurements. These data are consistent with earlier reports for heart rate (Cohen, 1980),  $VO_2$ ,  $VCO_2$ , VE and RER (Reilly and Brooks, 1990) during progressive exercise to maximal intensity. Recently, Deschenes et al. (1998) also reported that  $VO_2$ , VE and heart rate at the midpoint of progressive maximal exercise protocols show no time of day variations for 0800, 1200, 1600 and 2000 hrs. However, the values of these variables at the threshold were significantly different between 2 methods for the same time of day in the present study. Chicharro et al. (1997) also investigated the relationship between LT and VT during a ramp protocol during a cycle ergometric test with 39 trained men. However, there is no data about the time period of day when the tests were performed in their study to compare with the present study. They found that mean values of VT and LT expressed as heart rate, work rate and  $VO_2$  were significantly different.

**Table 2.** Correlation coefficients (r) between ventilatory and lactate thresholds at different times of day.

Variable	09:00 h	14:00 h	19:00 h
Time (sec)	0.62	0.77*	0.70
HR (b.min <sup>-1</sup> )	0.90**	0.92**	0.94**
VE (L.min <sup>-1</sup> )	0.71	0.74	0.88**
RER	0.59	0.69	0.56
$VO_2$ (ml.min <sup>-1</sup> )	0.88**	0.82*	0.89**
% $VO_{2max}$	0.81*	0.85*	0.76*

\*  $p < 0.05$ , \*\*  $p < 0.01$ .

HR= heart rate, VE= minute ventilation, RER= respiratory exchange ratio, % $VO_{2max}$ =  $VO_2$  relative to  $VO_2$  max.

The results of present study presented in Table 1 also indicate that blood lactate concentration increases before the rise in VE and  $VO_2$  during the incremental exercise for all time periods used,

leading us to suggest that ventilatory response to the exercise was triggered by blood lactate accumulation. It also indicates that time of day has no influence on this process.

The strength of the correlation coefficients between LT and VT observed in the present study were lower than the level observed in previous studies for  $VO_2$  ( $r=0.82-0.89$  in the present study versus  $r=0.94$  in the study of Reinhard et al., 1979), and for % $VO_2$  ( $r=0.76-0.85$  in the present study versus  $r=0.95$  in the study of Davis et al., 1976). The differences between the studies may be related to testing protocols, subjects' activity background or site of blood sampling.

## CONCLUSION

From the results of this investigation it appears that; 1) the values of the selected variables related to the threshold calculated by two methods were not influenced by time of day, 2) the strength of the relationship between VT and LT was high for heart rate and  $VO_2$  and the strength of the relationship was not influenced by time of day, 3) the values of the selected variables were significantly different between VT and LT for all time periods used in the study.

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