

RELATIONSHIP OF ERGOMETER-SPECIFIC $\dot{V}O_2$ MAX AND MUSCLE ENZYMES TO BLOOD LACTATE DURING SUBMAXIMAL EXERCISE

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ABSTRACT

This study compared the relationship of maximum oxygen uptake and skeletal muscle enzyme activities to the submaximal exercise intensity eliciting 4 mM blood lactate (OBLA). Twelve subjects performed both cycle (Cy) and treadmill (Tr) submaximal exercise with step-wise increments each fourth minute. Blood lactate concentration and oxygen uptake ($\dot{V}O_2$) were determined during the final minute of each step. Peak $\dot{V}O_2$ during exhaustive exercise was also measured on each ergometer. Biopsies were taken from the gastrocnemius (gast) and vastus lateralis (vl) muscles as representatives of muscles recruited during Tr and Cy exercise, respectively. Citrate synthase (CS), phosphofructokinase (PFK), and lactate dehydrogenase (LDH) activities were assayed. Peak $\dot{V}O_2$ was 10% greater and the $\dot{V}O_2$ at OBLA was 16% greater during Tr compared to Cy exercise. The percent of peak $\dot{V}O_2$ at OBLA was 85% and 79% for Tr and Cy exercise, respectively. The absolute enzyme activities were not different in the two muscles, however the ratio LDH/CS was greater in the vl than in the gast. The results indicate that the absolute differences between Cy and Tr exercise in peak $\dot{V}O_2$ are not commensurate with the differences in the relative exercise intensity at which OBLA occurs.

Keywords: Metabolism, Anaerobic threshold, Oxygen uptake.

INTRODUCTION

Exercise testing in the laboratory has traditionally included the determination of maximal oxygen uptake ($\dot{V}O_2$ max) as an indication of the capacity for endurance exercise (Saltin and Åstrand, 1967). The cycle (Cy) and the treadmill (Tr) are the two ergometers most commonly used for this purpose. Recently, maximal endurance exercise performance has been shown to be closely related to physiological measurements during submaximal exercise. In particular, it has been repeatedly demonstrated that an individual's endurance is correlated more strongly with the blood lactate response to submaximal exercise than with $\dot{V}O_2$ max (Farrell et al, 1979; Kumagai et al, 1982; Hagberg and Coyle, 1983). In light of the increasing popularity of using blood lactate related variables in the evaluation of aerobic fitness, the present study was designed to compare the blood lactate response to continuous, progressive exercise on the Cy and Tr ergometers. Specifically, the exercise intensities, both the absolute oxygen uptake ($\dot{V}O_2$) and $\dot{V}O_2$ relative to $\dot{V}O_2$ max, which corresponded to a blood lactate concentration of 4 mM (OBLA = onset of blood lactate accumulation) on each ergometer, were compared. It was hypothesised that any observed difference in this lactate determined intensity would be related to and commensurate with the difference in $\dot{V}O_2$ max measured between the two ergometers.

It has been previously shown that OBLA, in addition to being strongly correlated with marathon running performance (Sjodin and Jacobs, 1981), is also related to several metabolic characteristics of the muscle cell such as fibre type, capillary density, and key enzyme activities (Sjodin and Jacobs, 1981; Sjodin et al, 1981). Thus, it was also speculated that any differences in OBLA between Cy and Tr exercise would be reflected in differences in enzyme activities between the muscles chosen to be representative of the predominant muscle groups recruited during Cy or Tr exercise, i.e. the vastus lateralis and lateral gastrocnemius, respectively.

METHODS

The details of the experimental protocol and all associated risks or discomforts were explained to volunteers before obtaining their informed consent to participate as subjects. The protocol was approved by the Karolinska Hospital's Human Ethics Committee. The subjects were 12 males with a mean (\pm SD) age, height and weight of 23 ± 5 years, 180 ± 6 cm, and 75 ± 9 kg, respectively.

Muscle sampling. The subjects visited the laboratory on three occasions. On the first occasion two muscle samples were obtained with the needle biopsy technique (Bergström, 1962) from both the vastus lateralis and the lateral head of the gastrocnemius. One tissue sample from each muscle was mounted in an embedding medium and frozen in isopentane cooled with liquid nitrogen for later histochemical analyses. The second sample, for biochemical assays, was frozen directly in liquid nitrogen. The tissues were stored at -80°C until analysed. Muscle fibres were classified as slow twitch (ST) or fast twitch (FT) after staining for myofibrillar ATPase activity at a pH of 9.4, following preincubation at pH 10.3 (Guth and Samaha, 1970). Photographs of serial sections stained for NADH-tetrazolium reductase (Novikoff, 1961) were used to calculate the mean fibre area and the relative area occupied by each fibre type (Thorstensson, 1976; Tesch, 1980). The second tissue specimen from each muscle was freeze dried. Remaining visible impurities, connective tissue and blood clottings were dissected out before weighing the tissue on a Cahn® electrobalance. The tissue was then homogenised by sonication in an ice-cooled medium of 0.1 M phosphate buffer, pH 7.3. The activities of phosphofructokinase (PFK, EC 2.7.1.11), lactate dehydrogenase (LDH, EC 1.1.1.27), and citrate synthase (CS, EC 4.1.3.7) were determined with assays based on fluorometric techniques (Essén et al, 1980).

Submaximal exercise. Since most subjects cannot attain the same value for $\dot{V}O_2$ max during both Cy and Tr exercise, we will refer to the ergometer-specific $\dot{V}O_2$ max as "peak $\dot{V}O_2$ ". Cycle ergometer or treadmill tests were performed one and two weeks after the biopsies, with the order of testing being counter-balanced among subjects. Submaximal and maximal test protocols were administered for each ergometer. A warm-up was performed for 5 minutes on the Tr at an intensity estimated to require between 40 to 50% of peak $\dot{V}O_2$. Both submaximal exercise protocols were continuous with intensity increments every fourth minute of 50 W for the Cy and 0.5 to 2 $\text{km}\cdot\text{h}^{-1}$ for the Tr. The initial exercise intensity was 50 W on the Cy and 12 to 14 $\text{km}\cdot\text{h}^{-1}$ on the Tr, which was 0.5 to

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1 km.h⁻¹ faster than the warm-up velocity. Exercise was halted when subjects rated their perceived leg exertion during Cy exercise, or whole body exertion during running, as greater than six on the 10-point scale of ratings of perceived exertion (RPE) developed by Borg (1982). It has been previously reported that almost all subjects accumulate blood lactate concentrations greater than 4 mM at this RPE (Jacobs, 1981; Noble et al, 1983). Expired air was collected in Douglas bags during the final minute at each intensity and transferred into a wet spirometer for volume determination. The fractions of CO₂ and O₂ were analysed with a mass spectrometer (Centronics® MGA200) to enable the calculation of $\dot{V}O_2$. Blood samples (25 μ l) were taken from the finger-tip while the subject continued to exercise during the final 30 s at each intensity. The blood was deproteinised immediately and stored under refrigeration until assayed for lactate concentration with a fluorometric enzymatic technique (Karlsson et al, 1983). By plotting the relationship of lactate to exercise intensity for each subject, OBLA was interpolated as previously described (Jacobs, 1981).

Maximal exercise. A 30 min uncontrolled recovery period followed each submaximal exercise test after which peak $\dot{V}O_2$ was determined with a continuous, progressive protocol until voluntary exhaustion. For the Tr peak $\dot{V}O_2$ determination, running speed remained constant while slope increased stepwise by 0.5 degrees. 30 s⁻¹. For the Cy peak $\dot{V}O_2$ determination, exercise commenced at 100-250 W and increased stepwise by 50 W every two minutes until voluntary exhaustion. Blood was sampled for lactate determination four minutes after the maximal exercise tests.

Statistics. The BMDP Statistical Software package was used for all calculations (Dixon and Brown, 1977). Student's t-test for paired observations was used to determine the significance of differences between mean values. Statistical significance was set at the 0.05 level.

RESULTS

Maximal Exercise. Peak $\dot{V}O_2$ was higher during Tr exercise (mean \pm SD, 66 \pm 8 ml.kg⁻¹ min⁻¹) than during Cy exercise (60 \pm 6 ml.kg⁻¹ min⁻¹) ($p = 0.007$). There was a moderate but significant correlation between individual peak $\dot{V}O_2$ values on the two ergometers with 10 of the 12 subjects demonstrating a higher peak $\dot{V}O_2$ on the Tr ($r = 0.61$, $p < 0.05$) (Fig. 1). During Tr exercise the "levelling-off" criterion for $\dot{V}O_2$ was observed in nine of the 12 subjects since $\dot{V}O_2$ decreased, remained the same, or increased by less than 2 ml.kg⁻¹ min⁻¹ during the last 30 s of exercise in spite of the increased Tr elevations. Using the same criteria "levelling-off" was observed in only seven subjects during the maximal Cy exercise. All subjects stated that leg fatigue limited their ability to continue

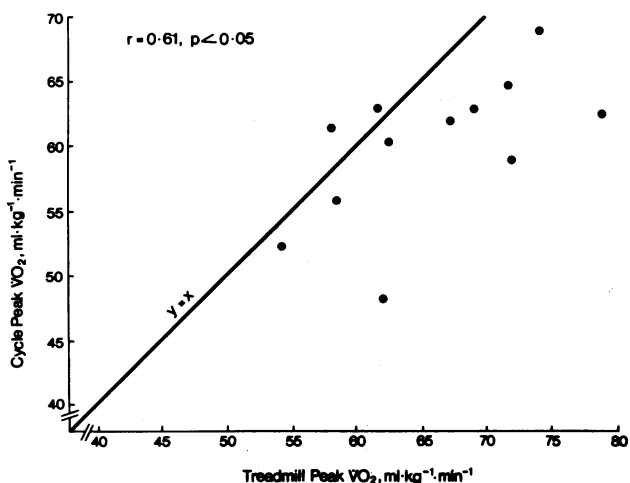


Fig. 1: The relationship of peak $\dot{V}O_2$ measured during exhaustive exercise on the treadmill to peak $\dot{V}O_2$ measured on the cycle.

Cy exercise. Peak blood lactate levels four minutes after the maximal tests were 9.8 \pm 1.6 and 10.3 \pm 1.2 mM for Tr and Cy exercise, respectively, and these were not significantly different.

Submaximal Exercise. The relationship between blood lactate and steady state $\dot{V}O_2$ is depicted in Figure 2.

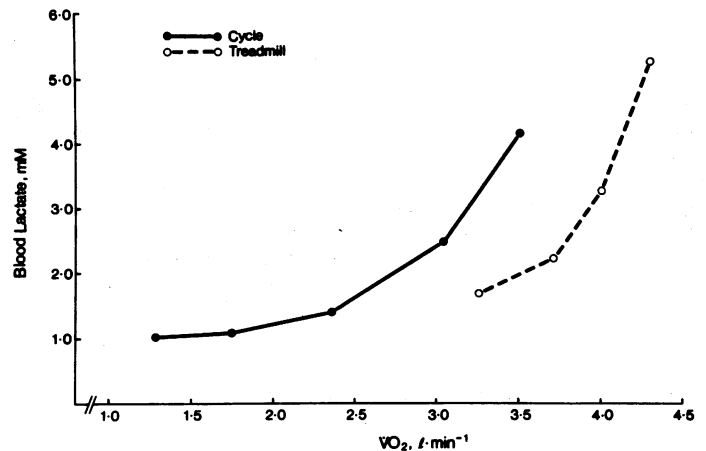


Fig. 2: Relationship of blood lactate to oxygen uptake during submaximal cycle and treadmill exercise.

Using OBLA as a reference point, $\dot{V}O_2$ was 16% higher during Tr than Cy exercise ($p < 0.001$). The relative utilisation of peak $\dot{V}O_2$ (% peak $\dot{V}O_2$) at OBLA was 85% Tr peak $\dot{V}O_2$ and 79% Cy peak $\dot{V}O_2$ ($p < 0.03$) (Fig. 3, Table 1).

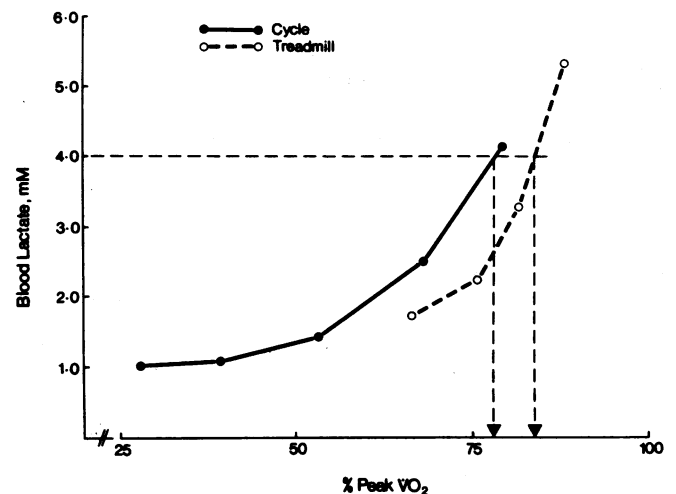


Fig. 3: Relationship of blood lactate to the relative utilisation of peak $\dot{V}O_2$ during submaximal cycle and treadmill exercise. The arrows indicate the % peak $\dot{V}O_2$ at 4 mM lactate.

Table 1 Peak $\dot{V}O_2$ during maximal exercise, the absolute and relative $\dot{V}O_2$ corresponding to 4mM blood lactate (OBLA).

	Peak $\dot{V}O_2$ ml.kg ⁻¹ min ⁻¹	$\dot{V}O_2$ OBLA ml.kg ⁻¹ min ⁻¹	$\dot{V}O_2$ OBLA % peak $\dot{V}O_2$
Treadmill	66 \pm 8	57 \pm 5	85 \pm 5
Cycle	60 \pm 6	48 \pm 6	79 \pm 7
% difference	9	16	7
P	0.007	0.001	0.024

Values are expressed as mean \pm SD. $p =$ significance of difference between means.

The $\dot{V}O_2$ at OBLA was not significantly correlated to peak $\dot{V}O_2$ on either ergometer. Nonsignificant inter-ergometer correlation coefficients were observed for the $\dot{V}O_2$ at OBLA, the % peak $\dot{V}O_2$ at OBLA, and the absolute exercise intensity at OBLA (i.e. W.kg⁻¹ for the Cy vs. m.s⁻¹ for the Tr).

Muscle characteristics. The frequency of ST fibres in the two muscles sampled is shown in Figure 4 and the mean values did not differ significantly.

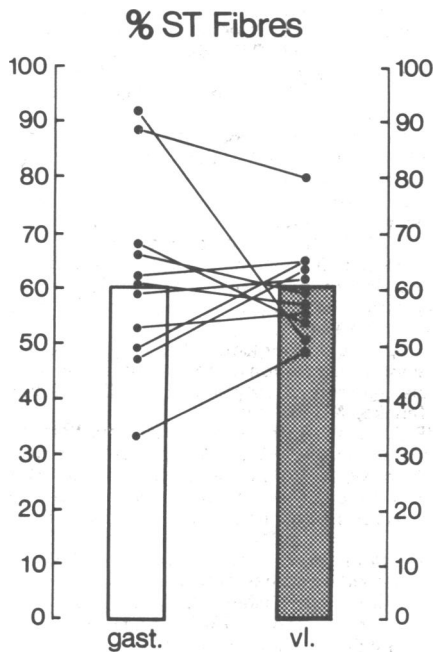


Fig. 4: Frequency of slow twitch (ST) fibres in the *m. vastus lateralis* (vl) and lateral head of the *m. gastrocnemius* (gast). The staples represent the mean values.

The mean FT and ST fibre areas were 5511 ± 1556 and $5797 \pm 1157 \mu\text{m}^2$, respectively, in the vastus lateralis, and 6249 ± 2135 and $5318 \pm 1647 \mu\text{m}^2$ in the gastrocnemius. The differences in mean fibre area between muscles were not significant nor was the percent muscle area occupied by a given fibre type. Not all enzyme assays could be performed for both muscles of four of the subjects due to the small biopsy specimen size and their enzyme data has been excluded from the statistical analyses. The two muscles did not differ in the absolute activity levels of CS, PFK, and LDH, but the ratio of the glycolytic enzymes, LDH or PFK, to the oxidative enzyme marker, CS, did differ significantly. The vastus lateralis demonstrated a higher ratio suggesting a higher glycolytic relative to oxidative activity potential than the gastrocnemius (Fig. 5).

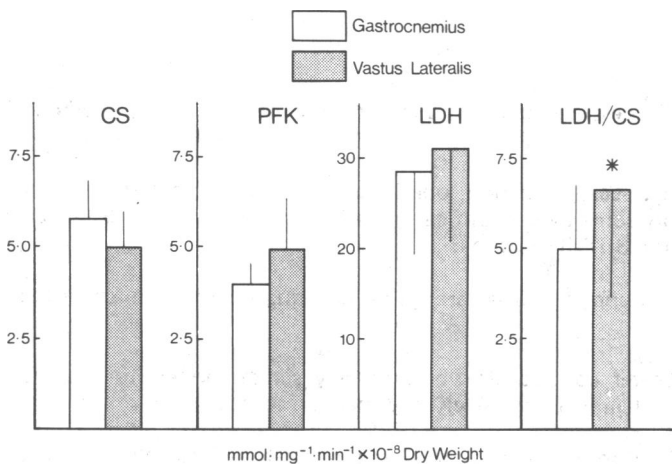


Fig. 5: Activities of citrate synthase (CS), phosphofruktokinase (PFK), lactate dehydrogenase (LDH), and the LDH/CS ratio in the gastrocnemius and vastus lateralis. Staples represent mean and SD.

The difference between the ergometers in either the absolute VO_2 or % peak VO_2 at OBLA was not significantly related to the absolute difference between ergometers in the measured peak VO_2 .

DISCUSSION

The present study has clearly demonstrated that a blood lactate concentration of 4 mM occurs at a higher steady state VO_2 during Tr running than during Cy exercise. This is consistent with several earlier investigations (Hermansen and Saltin, 1969; Koyal et al, 1976; Kindermann et al, 1980).

The 9% greater peak VO_2 during Tr exercise is similar to previous investigations (Hermansen and Saltin, 1969; Kindermann et al, 1980). This higher Tr peak VO_2 has been attributed to a larger stroke volume leading to a greater cardiac output when compared to maximal Cy exercise (Hermansen and Saltin, 1969; Hermansen et al, 1970). It was hypothesised that these functional differences would affect OBLA to the same extent that they affect the oxygen transport system. The end result would be a similar % peak VO_2 corresponding to a given lactate concentration during both forms of exercise. If such were the case the higher lactate level at a given absolute VO_2 during Cy exercise could possibly be explained by the assumed smaller muscle mass being required to perform the same work as during Tr exercise. This, however, was not the case. The relative difference between ergometers in the VO_2 at OBLA was almost twice as great as the difference in peak VO_2 (16% vs. 9%), similar to what was reported by Kindermann et al (1980). The end result was the significantly higher % peak VO_2 at OBLA during Tr exercise.

In order to evaluate the relationship of some metabolic characteristics of the exercising muscle to the inter-ergometer difference observed in the blood lactate responses the lateral head of the gastrocnemius was chosen to be representative of running, and the vastus lateralis to be representative of cycling. The choice may be questioned since the vastus lateralis is recruited also during running. However, muscle substrate depletion studies on biopsies taken from various leg muscles after Tr running indicate a far greater recruitment of the gastrocnemius (Costill et al, 1974). It should be emphasised that the gastrocnemius and the vastus lateralis are only considered to be representative of the predominant muscle groups involved in each type of exercise.

Although they did not present any experimental evidence, Kindermann et al (1980) suggested that a lower oxidative capacity of the exercising musculature during Cy exercise could account for the earlier onset of lactate accumulation during this form of exercise. This suggestion is supported in the present study where the activities of both glycolytic and oxidative "marker" enzymes have been assayed. Although the absolute enzyme activities did not differ between muscles, the ratio of the glycolytic to oxidative activities suggested a relatively greater oxidative potential in the gastrocnemius than in the vastus lateralis. Green et al (1981) also reported higher LDH activities in the vastus lateralis than in the gastrocnemius, although oxidative enzyme activities were similar in the two muscles. The theoretical metabolic consequence is a greater capacity to oxidise pyruvate through oxidative phosphorylation and consequently a delayed production and accumulation of lactate in muscle and blood. This ratio has been shown previously to be directly related to the exercise intensity at OBLA (Sjödín et al, 1981).

It still remains to be demonstrated that the higher blood lactate concentrations at a given submaximal VO_2 during Cy exercise are reflective of greater intramuscular lactate production when compared to the same VO_2 during Tr exercise. An alternative explanation of the "delayed" accumulation of blood lactate during a specific form of exercise may revolve around the rate of lactate clearance from muscle and blood (Donovan and Brooks, 1983). It may be speculated that Tr running is associated with a larger active muscle mass, greater capillarisation and blood flow in the active muscles, and a metabolic profile in the exercising musculature which favours

lactate oxidation and/or gluconeogenesis from lactate to a greater extent than the musculature involved during Cy exercise. All of these factors could directly affect the rate of lactate clearance from muscle tissue and blood.

In conclusion, the steady state $\dot{V}O_2$ at OBLA was significantly higher during Tr than during Cy exercise. This difference was greater than could be accounted for by the difference between exercise modes in peak $\dot{V}O_2$. Consequently, OBLA occurred at a higher % peak $\dot{V}O_2$ during Tr running than during Cy exercise. There are indications that this difference may be due, at least partially, to differences in the metabolic profile of the exercising musculature, as reflected by the ratio of key oxidative to glycolytic enzyme activities, in addition to the established differences in certain cardiovascular variables associated with Tr and Cy exercise.

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BOOK REVIEW

Title: HEALTH AND PHYSICAL FITNESS
Author: William P. Marley, PhD, FACSM
Publishers: Saunders College Publishing, Philadelphia &c., 1982
 412 pages Paperback

This book was written for a particular college course in Health and Fitness Foundations and confines itself to basic principles of the main subjects in this field.

The first part, on health is well set out and covers coronary heart disease, nutrition, obesity and weight control before moving on to positive health factors and exercise regimes. There are inevitably some oversimplifications which will irritate medical readers and the orientation of the book is very American, which may irritate the British PE reader.

While the health and risk factors are well covered, the sections on sports injuries are quite inadequate, not so much in their accuracy, but in the brief and eclectic coverage of the subject.

The health sections are well worth reading, the fitness part is better covered elsewhere in very similar books (for instance, Sharkey's *Physiology of Fitness*) and there is a full bibliography, as usual almost exclusively to American sources.

This book is more readable than many and may well serve a useful purpose in its chosen context of health education for a mixed audience, but its inherent limitations make it a first port of call, not a definitive text.

P. N. Sperry