



## Lactate Threshold (D-Max Method) and Maximal Lactate Steady State in Cyclists

by

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*The Maximal Lactate Steady State (MLSS) is defined as the highest workload that can be maintained over time where there is a balance between lactate production and lactate clearance. Therefore, determination of this workload is very importance for diagnosis of aerobic capacity and training program design.*

*The main objective of this study was to evaluate the validity of lactate threshold values determined by the D-max method as related to MLSS in cyclists. The research material included 10 female (body height 167±5.7 cm; body mass 56±4.8 kg; percent body fat 12.3±2.1) and 10 male well-trained cyclists (body height 183.5±4.4 cm; body mass 73.2±4.1 kg; percent body fat 7.9±2.6). The research had two distinct phases, separated by one day of active recovery. During the first phase, progressive tests were carried out to determine lactate threshold and maximal oxygen uptake in each subject. During the second phase, each athlete performed a series of 30-min ergocycle tests, with a fixed workload to establish maximal lactate steady state.*

*Results showed no significant differences between lactate threshold workload (WRLT), determined by the D-max method, and maximal lactate steady state workload (WRMLSS) in female and male cyclists, expressed in absolute and relative values. Differences between male and female cyclists in absolute and relative values of WRLT, WRMLSS, and WRmax were significant ( $p<0.05$ ), but in relative values there was a tendency for decreased differences between groups. The oxygen uptake at the lactate threshold and MLSS were significantly ( $p<0.05$ ) different. Also, a significant ( $p<0.05$ ) difference was observed in values of heart rate and lactate concentration at the lactate threshold and MLSS. The analysis of changes in lactate concentration, heart rate and oxygen uptake between the 10th and 30th minutes of MLSS, indicates that there was a significant ( $p<0.05$ ) increase in these values in male and female cyclists. The strong correlation ( $r=0.97$ ;  $p<0.05$ ) between WRLT and WRMLSS was found. Also, a significant correlation between ( $r=0.96$ ;  $p<0.05$ ) WRMLSS and peak workload during the incremental test (WRmax) ( $r=0.96$ ;  $p<0.05$ ) was also observed.*

**Key words:** cyclists, lactate threshold, maximal lactate steady state

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## Introduction

For almost 50 years now, the blood lactate curve and metabolic thresholds have become standard in the diagnosis of aerobic capacity and endurance performance in athletes. During this time, there were many debates concerning the methodology and physiological background used in determination of the anaerobic threshold (AT). In the last few years, the maximal lactate steady state (MLSS) has been proposed as a valid and reliable marker of endurance performance (Snyder et al., 1994).

The Maximal Lactate Steady State (MLSS) is defined as the highest workload that can be maintained over time without continued blood lactate accumulation. By definition, MLSS is attained when lactate (LA) varies by less than a 1 mM during the final 20 min of constant intensity exercise. The physiological importance of the MLSS workload is that it defines the exercise intensity above which there is a net contribution of energy associated with lactate accumulation, due to an increased rate of glycolysis that exceeds the rate of mitochondrial pyruvate utilization (Heck et al., 1985; Bkneke et al., 1996). During the work efforts at and below the MLSS workload, there is a balance between lactate production and its removal, but when the effort is above the MLSS workload, the rate of lactate production exceeds its rate of clearance.

The MLSS workload intensity seems to be the highest value when stability in the acid-base balance and pulmonary gas exchange are still observed (Gresser et al., 1996). For this reason, direct determination of MLSS is considered a gold standard for evaluating endurance performance in athletes, which can make endurance training more effective.

The determination of the MLSS workload has a serious disadvantage, because it requires the performance of a series of constant-intensity tests for each athlete. Such testing procedures require the presence of an athlete in the laboratory for several days, which can have a detrimental effect on the training program. To eliminate this problem, some researches try to determine the MLSS indirectly, based on a single incremental test (Heck et al., 1985; Jones et al., 1988; Poole, 1988; Beneke, 1996; McLellan, 1992; Laplaud et al., 2005; Figueira et al., 2008) and on field tests (Hoogeveen et al., 1997; Harnish et al., 2001).

Current research results of indirect determination of MLSS are controversial. Some studies confirm that the MLSS can be estimated with a single exercise test, but others observed a relatively wide individual variability. There is also no information about how the lactate threshold, determined by the D-max method, corresponds to the maximal lactate steady state (MLSS).

Therefore, the main objective of the present investigation was to determine the validity of the lactate threshold, evaluated by the D-max method, as proposed by Cheng et al. (1992), when evaluating the MLSS in cyclists.

## Material and methods

The research material (mean $\pm$ SD) included 10 female (body height 167 $\pm$ 5.7 cm; body mass 56 $\pm$ 4.8 kg; percent body fat 12.3 $\pm$ 2.1) and 10 male, well-trained cyclists (body height 183.5 $\pm$ 4.4 cm; body mass 73.2 $\pm$ 4.1 kg; percent body fat 7.9 $\pm$ 2.6). All tested subjects possessed current medical examinations, confirming proper health status and the ability to perform exhaustive exercise. The research project was approved by the Ethics Committee for Scientific Research at the Jerzy Kukuczka Academy of Physical Education in Katowice.

The research had two phases, separated by one day of active recovery. At the beginning of the first phase the values of body height, mass and body composition (FFM, Fat%, TBW) were evaluated. For analysis of body composition, the electrical impedance method (InBody 220, Biospace) was used. Following, the incremental tests were carried out to determine lactate threshold and maximal oxygen uptake. The tests were performed on an ergocycle (Excalibur Sport, Lode), beginning with a workload of 30W for female and 40W for male athletes. Workload was then increased by that starting value every 3 min until volitional exhaustion. During the exercise protocol, the following variables were constantly registered: heart rate (HR), minute ventilation (VE), oxygen uptake (VO<sub>2</sub>) and expired carbon dioxide (CO<sub>2</sub>) (MetaLyzer 3B-2R, Cortex). At the beginning and the end of each work load, capillary blood samples were drawn to determine lactate concentration. These values allowed for determining the lactate thresholds, by the D-max method, as proposed by Cheng et al. (1992), for each athlete. The D-max is defined as the point on the regression curve that yields the maximal distance to the straight line formed by

Table 1

*The incremental and MLSS test results in male and female cyclists*

	Male Group			Female Group		
	Mean±SD	Min-Max	Significant difference	mean±SD	Min-Max	Significant difference
WR <sub>max</sub> (W)	400±24,1	360-440	NS	282±32,2	240-330	NS
WR <sub>LT</sub> (W)	281,6±24,4	240-320		192±32,2	150-240	
WR <sub>MLSS</sub> (W)	288,3±19,4	260-320		195±28,2	165-240	
VO <sub>2 max</sub> (ml/kg/min)	69,6±6,2	60-80		57,8±4	54-63	
VO <sub>2 LT</sub> (ml/kg/min)	54,5±4,2	49-62	*	44,2±5,5	35-50	*
VO <sub>2 MLSS</sub> (ml/kg/min)	57,7±4,1	52-65		47,2±4,1	41-53	
HR <sub>max</sub> (bpm)	192±9,8	180-204		188,8±5,9	180-197	
HR <sub>LT</sub> (bpm)	170,8±11,7	155-188	*	167,2±7,2	160-180	*
HR <sub>MLSS</sub> (bpm)	181,1±8,03	165-187		178,4±6,6	171-185	
LA <sub>max</sub> (mmol/l)	9,24±1,5	7,6-12,1		8,6±0,5	8-9,16	
LA <sub>LT</sub> (mmol/l)	2,7±0,3	2,2-3,1	*	2,6±0,1	2,3-2,8	*
LA <sub>MLSS</sub> (mmol/l)	4,8±0,69	3,6-5,5		4,2±0,2	3,8-4,3	
LA <sub>12'recovery</sub> (mmol/l)	2,1±0,7	1,1-2,9		2,8±0,9	1,3-4,1	

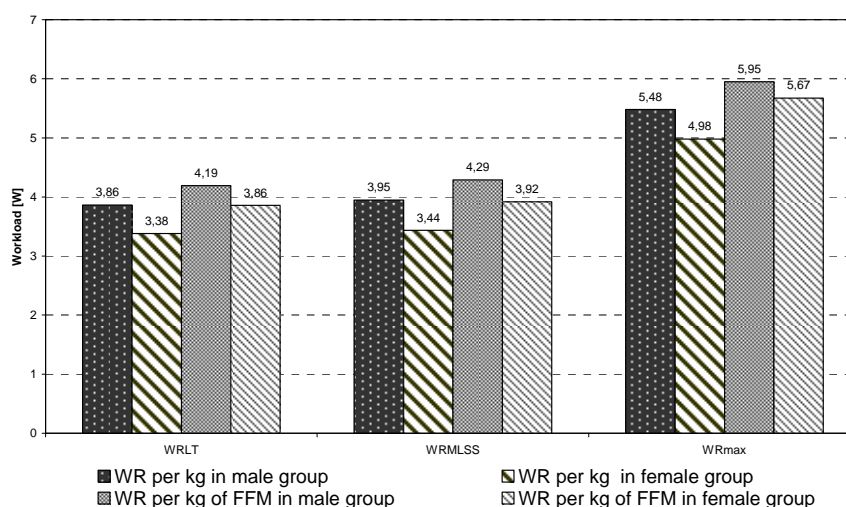
\**p*<0.05; NS – non-significant

the two end points of the curve (lactate concentration at rest and after the final stage of the incremental test). Capillary blood samples were also drawn after the 3<sup>rd</sup>, 6<sup>th</sup>, 9<sup>th</sup> and 12<sup>th</sup> min after cessation of the test to determine lactate utilization.

During the second phase, each athlete performed a series of 30-min ergocycle tests, with a fixed workload, to establish maximal lactate steady state. Every test was preceded by a 10-min warm-up ride with the resistance set at 65-70% of the lactate threshold workload (second zone – endurance training), with controlled individual pedal frequency. Capillary blood samples were drawn before

and after the warm-up ride, and every 5-min of the 30-min test (5, 10, 15, 20, 25, and 30 min) to determine lactate concentration. Each athlete began the series of tests with an individual workload at lactate threshold (D-max method). When lactate concentration did not rise, more than 1 mmol/l in the final 20 minutes of the test, the test was repeated once again after 48h of rest with higher workloads (15W for female and 20 W for male).

The obtained data was analyzed statistically with the use of Statistica 8.0. The results were presented as arithmetic means (x) and standard deviations (SD). To determine the significance of differences



**Fig. 1** Average values of relative workload (WR) at the lactate threshold (LT), and maximal lactate steady state (MLSS) in male and female cyclists

Table 2

The values of lactate concentration, heart rate and their changes during the MLSS test in male and female cyclists

	Male Group		Female Group	
	mean±SD	Min-Max	mean±SD	Min-Max
LA <sub>5'</sub> MLSS (mmol/l)	3,2±0,4	2,7-3,7	3,1±0,1	2,8-3,2
LA <sub>10'</sub> MLSS (mmol/l)	3,9±0,6	2,9-4,6	3,4±0,1	3,2-3,4
LA <sub>15'</sub> MLSS (mmol/l)	4,2±0,6	3,2-4,7	3,6±0,1	3,5-3,8
LA <sub>20'</sub> MLSS (mmol/l)	4,4±0,6	3,3-4,9	3,8±0,1	3,6-4,1
LA <sub>25'</sub> MLSS (mmol/l)	4,5±0,6	3,4-5,1	4,1±0,2	3,6-4,2
LA <sub>30'</sub> MLSS (mmol/l)	4,8±0,6	3,6-5,5	4,2±0,2	3,8-4,3
ΔLA <sub>10'-30'</sub> MLSS (mmol/l)	0,8±0,1	0,6-0,9	0,8±0,1	0,5-0,9
HR <sub>5'</sub> MLSS (bpm)	169±7,1	156-175	163±2,5	160-165
HR <sub>10'</sub> MLSS (bpm)	174±7,8	158-180	171±6,3	163-177
HR <sub>15'</sub> MLSS (bpm)	176±7,8	160-182	173±7,5	165-181
HR <sub>20'</sub> MLSS (bpm)	178±7,8	162-184	175±7,5	167-183
HR <sub>25'</sub> MLSS (bpm)	180±8,2	163-186	177±6,6	170-184
HR <sub>30'</sub> MLSS (bpm)	181±8,1	165-187	178±6,6	171-185
ΔHR <sub>10'-30'</sub> MLSS (bpm)	7,1±1,7	4-9	7,2±1,1	6-8
Δ%HR <sub>10'-30'</sub> MLSS (%)	4,1±1	2,3-5,2	4,4±0,4	3,6-4,9
Δ%VO <sub>2</sub> <sub>10'-30'</sub> MLSS (ml/kg/min)	2,4±0,9	1-4	3,1±0,8	2-4

between analyzed variables, the Wilcoxon's test was used. To determine the significance of differences between groups the U Mann-Whitney test was applied. The relationships between particular variables were determined by calculating the Spearman correlation coefficients. The level of statistical significance was set at  $p < 0.05$ .

## Results

The incremental and MLSS test results, as well as the significance of differences between variables, are presented in Table 1.

The statistical analysis indicate that there were no significant differences between lactate threshold workload ( $WR_{LT}$ ), determined by the D-max method, and maximal lactate steady state workload ( $WR_{MLSS}$ ) in female and male cyclists. This was also true for the relative values of workloads at LT and MLSS, calculated by body mass and body fat free mass (Figure 1). Differences between male and female cyclists in absolute and relative values of  $WR_{LT}$ ,  $WR_{MLSS}$ , and  $WR_{max}$  were significant ( $p < 0.05$ ), but in relative values of these variables there was a tendency for decreased differences between these groups. However, the oxygen uptake at the lactate threshold and MLSS were significantly ( $p < 0.05$ ) different, but the oxygen uptake was higher at the MLSS in both groups. Also, significant ( $p < 0.05$ ) differences were observed in values of heart rate and lactate concentration at the lactate threshold and MLSS. In both groups these values were higher at

the MLSS than at LT or with oxygen uptake (Table 2). The analysis of changes of lactate concentration, heart rate and oxygen uptake between the 10<sup>th</sup> and 30<sup>th</sup> minute of MLSS, indicates that there was a significant ( $p < 0.05$ ) increase in these values. The heart rate increased by  $4.1 \pm 1\%$  in males and  $4.4 \pm 0.4\%$  in female athletes (Table 2). The results of the U Mann-Whitney test indicate that there were no significant differences in percent of heart rate increase ( $\Delta\%HR_{10-30' MLSS}$ ) during the final 20 minutes of the MLSS test between male and female cyclists (Table 2).

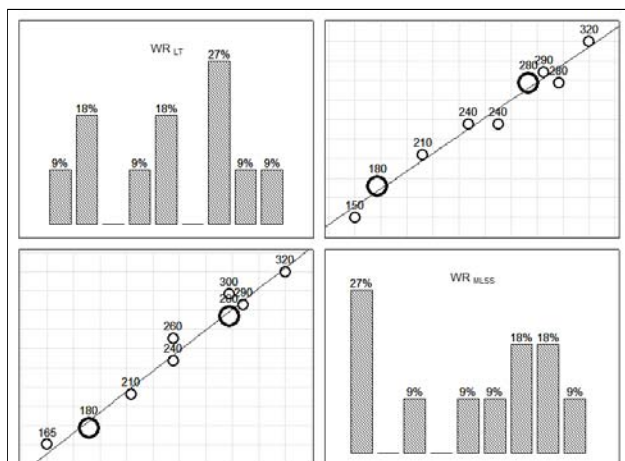
Correlation coefficients between analyzed variables and the LT and MLSS in the tested athletes are presented in Table 3.

The results indicate an almost full correlation ( $r = 0.97$ ;  $p < 0.05$ ) between lactate threshold workload ( $WR_{LT}$ ) and maximal lactate steady state workload ( $WR_{MLSS}$ ) (Figure 2), as well as between maximal lactate steady state workload ( $WR_{MLSS}$ ) and peak workload in the incremental test ( $WR_{max}$ ) ( $r = 0.96$ ;

Table 3

Correlation coefficients between chosen variables of the MLSS and LT in the tested athletes;  $p < 0.05$

Paired Variables	R
$WR_{MLSS}$ & $WR_{max}$	0,96
$WR_{MLSS}$ & $WR_{LT}$	0,97
$WR_{MLSS}$ & $VO_{2max}$	0,76
$WR_{LT}$ & $VO_{2max}$	0,81
$\Delta LA_{10-30' MLSS}$ & $\Delta HR_{10-30' MLSS}$	0,87
$\Delta LA_{10-30' MLSS}$ & $\Delta\%HR_{10-30' MLSS}$	0,61



**Fig. 2**

*The distribution of lactate threshold workload values and the maximal lactate steady-state workload in tested athletes*

$p < 0.05$ ). A positive, significant correlation ( $r = 0.87$ ;  $p < 0.05$ ) between the increase in lactate concentration ( $\Delta LA_{10-30' MLSS}$ ) and increase in heart rate ( $\Delta HR_{10-30' MLSS}$ ) during the final 20 minutes of the MLSS test were also observed. A strong, significant correlation ( $r = 0.61$ ;  $p < 0.05$ ) was also observed between the increase in lactate concentration ( $\Delta LA_{10-30' MLSS}$ ) and heart rate increase ( $\Delta \% HR_{10-30' MLSS}$ ) during the final 20 minutes of MLSS test.

## Discussion

Endurance refers to the length of time that an individual can perform work of a given intensity. The main factor which affects and limits this performance is fatigue. Even though a high maximal oxygen uptake ( $VO_{2max}$ ) is a prerequisite for success in endurance sports, the ability to sustain a high percent of  $VO_{2max}$  without accumulating fatigue is of greater importance. This is why the MLSS can be very a useful discriminator of endurance performance between athletes with similar maximal oxygen uptake values (Coyle et al., 1988). Jones et al. (1998) describe the MLSS as the highest predictive power of endurance performance from all measures of aerobic fitness. Beneke et al. (2000) observed that subjects with higher maximum performance in incremental tests have higher MLSS workloads. They found a positive correlation between  $WR_{max}$  and  $WR_{MLSS}$  ( $r = 0.82$ ;  $p < 0.001$ ). Our research, confirm these findings. We also observed a very strong correlation between  $WR_{MLSS}$  and  $WR_{max}$  ( $r = 0.96$ ;  $p < 0.05$ ) in tested cyclists. This data confirms that the MLSS is an excellent and very reliable marker of aerobic fitness, but the

determination of the MLSS is time consuming and requires great precision. This procedure requires the athlete to perform several tests of constant-intensity exercise on different days, which could seriously disturb the training program.

Therefore, the indirect determination of the MLSS can be a very useful instrument for a training program design. The pioneers in that field were Heck et al. (1985), who based the indirect determination of the MLSS workload on a single incremental test using a fixed 4 mM lactate threshold. Some studies confirm these findings (Jones et al., 1988; Poole, 1988), yet others observed a relatively wide individual variability (Stegmann et al., 1981).

Stegmann et al. (1981) observed that the absolute values of steady-state blood lactate concentration vary widely across individuals. Upon these findings, and the knowledge of the mechanism of lactate diffusion from muscle to blood, they proposed the concept of the individual anaerobic threshold (IAT). Kindermann et al. (1979) suggested that the IAT may be significantly better than a fixed blood lactate concentration in defining the MLSS. This concept has also been supported by other studies (Beneke, 1996; McLellan, 1992).

Beneke (1995), who evaluated the validity of the fixed lactate threshold at 4mmol/l (AT4) and IAT, as related to the MLSS during rowing ergometry, observed that AT4 and IAT do not represent the MLSS for the workload in rowing. In a different study, Beneke (2001) observed that the MLSS intra-individuality depends on the mode of exercise. The lactate concentration at the MLSS in rowing and cycling depends on the motor pattern of exercise. The MLSS seems to decrease with increasing mass of the primarily engaged muscle. This indicates that task-specific levels of MLSS occur at distinct levels of power output per unit of primarily engaged muscle mass.

The results of our study showed that the workload associated with a lactate threshold, as determined by the D-max method during an incremental test, was not different from that determined for the MLSS, for both female and male cyclists. Also, a very strong relationship between  $WR_{LT}$  and  $WR_{MLSS}$  ( $r = 0.97$ ;  $p < 0.05$ ) was found. However, statistical analysis showed a significant difference in lactate concentration, heart rate and oxygen uptake between LT and MLSS, and a significant increase in these variables during the final 20 minutes of the MLSS test.

The values of lactate concentration at LT and MLSS observed in our study were not much different than in other studies on cyclists (Beneke et al., 1996; Swensen et al., 1999; Lajoie et al., 2000). The significant ( $p < 0.05$ ) differences between lactate concentration at LT and MLSS, as well as a very high and significant ( $p < 0.05$ ) increase in values of this variable during the first 10 minutes of the MLSS effort, is related to lactate diffusion from the intramuscular compartment to plasma, which depends on the capacity of the monocarboxylic (MCT) transporters (Juel et al., 1994). This may explain the difference between lactate concentration in the LT and MLSS. The 3 min load increments applied in our exercise protocol, most likely failed to equilibrate lactate concentration, and its measurements in blood plasma at each workload may not yield the value of intramuscular lactate. This phenomenon may be confirmed by a significant post-exercise increase in lactate concentration observed after the progressive test. These findings were confirmed by McLellan et al. (1993), who observed that in protocols with load increments less than 5 min, lactate diffusion from the intramuscular compartment to plasma is insufficient and partly limited by the capacity of the MC transporters. Our results of lactate concentration during the MLSS showed that the length of increment for optimal lactate diffusion should be much longer than 5 min. We observed a significant increase in lactate concentration between 5-10 min of the MLSS effort. These findings are confirmed by Faxdal et al. (1994, 1996), who proposed an 8-min load increment for optimal lactate diffusion.

Increased HR during the final 20 minutes of the MLSS was significant in male ( $7.1 \pm 1.7$  bpm) and female ( $7.2 \pm 1.1$  bpm) cyclists. Such an increase, as in our study, was observed in other research using cycle ergometry (Snyder et al., 1994; Urhausen et al., 1993). Snyder et al. (1994) observed an average increase of 9 bpm in HR during the final 20 minutes of the MLSS. This data was also confirmed by Urhausen et al. (1993), who showed a 5 bpm increase in HR between the 15<sup>th</sup>-30<sup>th</sup> min of the MLSS. Previously, this HR drift during a fixed workload was associated with exercise-induced dehydration and

blood pooling in the lower extremities and skin, resulting in lower stroke volume (Rowell, 1993). However, dehydration and thermoregulation may not be the only causes in which HR increases during continuous exercise. There is also some evidence that increases in catecholamine levels, associated with rises in body temperature,  $H^+$  and lactate concentration, may cause a rise in the heart rate during prolonged exercise (Rowland, 2005). In our research, we found a significant relationship ( $r = 0.87$ ;  $p < 0.05$ ) between the increase in lactate concentration ( $\Delta LA_{10-30' MLSS}$ ) and heart rate ( $\Delta HR_{10-30' MLSS}$ ) during the final 20 minutes of the MLSS. We believe that heart rate variability is mainly related to the changes in circulating catecholamine concentration, which may reflect the sympathetic control of heart rate and metabolism. This reaction could also be partially caused by a decrease in muscle efficiency, as a result of fatigue. Lajoie et al. (2000), like others (Swensen et al. 1999), reported a significant increase in oxygen uptake ( $VO_2$ ) during exercise at the MLSS. Our research confirms this finding, as we observed a significant ( $p < 0.05$ ) increase in  $VO_2$  registered between 10<sup>th</sup>-30<sup>th</sup> minutes of exercise at the MLSS. This increase in  $VO_2$  could reflect a drop in muscle economy during exercise, likely caused by a progressive recruitment of type II muscle fibers with increasing exercise duration; thereby, resulting in a gradual fatigue of previously recruited type I muscle fibers (Cole et al., 1992).

### *Practical Applications*

The lactate threshold determined by the D-max method can be used to estimate the MLSS in cyclists. A strong relationship between the MLSS workload and the maximal workload in the incremental test observed in the present study, suggests that the MLSS workload is a useful predictor of aerobic capacity. Also, a strong relationship between the increase in lactate concentration, heart rate and the percent of heart rate increase during the final 20 minutes of the MLSS test, suggests that the analysis of heart rate drift during a fixed workload can be used for indirect determination of the MLSS.

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