

Time of VO₂max plateau and post-exercise oxygen consumption during incremental exercise testing in young mountain bike and road cyclists

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Short title:

VO₂max plateau during incremental exercise in cyclists

Summary

The purpose of this study was to compare markers of glycolytic metabolism in response to the Wingate test and the incremental test in road and mountain bike cyclists, who not different performance level and aerobic capacity. All cyclists executed the Wingate test and incremental test on a cycle ergometer. Maximal power and average power were determined during the Wingate test. During the incremental test the load was increased by 50 W every 3 min, until volitional exhaustion and maximal aerobic power (AP_{max}), maximal oxygen uptake (VO_{2max}), and time of VO_{2max} plateau (T_{plateau}) were determined. Post-exercise measures of oxygen uptake (VO_{2post}), carbon dioxide excretion, (VCO_{2post}), and the ratio between VCO₂/VO₂ (RER_{post}) were collected for 3 min immediately after incremental test completion. Arterialized capillary blood was drawn to measure lactate (La⁻) and hydrogen (H⁺) ion concentrations in 3 min after each test. The data demonstrated significant differences between mountain bike and road cyclists for T_{plateau}, VO_{2post}, VCO_{2post}, La⁻ which was higher-, and RER_{post} which was lower-, in mountain bike cyclists compare with road cyclists. No differences were observed between mountain bike and road cyclists for AP_{max}, VO_{2max}, H⁺ and parameters measured in the Wingate test. Increased time of VO_{2max} plateau concomitant to larger post-exercise La⁻ and VO₂ values suggests greater anaerobic contribution during incremental testing efforts by mountain bike cyclists compared with road cyclists.

Key words: time of maximal oxygen plateau, anaerobic metabolism, cycling

Introduction

Performance in the cross-country Olympics (XCO) subdiscipline of mountain biking (MTB) is commonly accredited to aerobic capacity (Smekal et al. 2015) and terrain handling

skills (Impellizzeri and Marcora 2007). Regarding the former, the main physiological constituents of aerobic performance are power produced at the ventilatory threshold, maximal aerobic power, and maximal oxygen uptake relative to body mass (Gregory et al. 2007; Impellizzeri and Marcora 2007; Impellizzeri et al. 2005; Smekal et al. 2015). The same discriminative indicators are considered crucial in road cycling, which is also performed at variable intensities over extended periods of time (Mujika and Padilla 2001; Wilber et al. 1997). However, competitive road cycling is quite distinctive from MTB even when performed in mountainous or hilly terrain. Road races are considerably longer in duration and often consist of multiple stages. Race in mountain biking is performed entirely off-road, requiring significant isometric muscle work (Impellizzeri and Marcora 2007) and considerable balance to circumnavigate the rough features of a course (Hebisz et al. 2014). Furthermore, mountain biking competition involves highly variable power demands, with activity frequently performed at high and maximal levels of heart rate, oxygen uptake, and blood lactate concentrations (approximately 6 mmol/l) (Gregory et al. 2007; Impellizzeri et al. 2002; Smekal et al. 2015; Stapelfeldt et al. 2004). Hence, the specificity of each cycling discipline suggests differences in those physiological and performance characteristics that relate to performance.

Macdermid and Stannard (2012) quantified the power output of a simulated mountain biking race to find that highest power was produced immediately after the start and during uphill segments, incurring a significant oxygen deficit. Therefore, the high-intensity yet intermittent nature of MTB racing has led to suggestions that anaerobic metabolism is just as an important determinant of performance as aerobic metabolism (Impellizzeri and Marcora 2007; Macdermid and Stannard 2012; Stapelfeldt et al. 2004). Inoue et al. (2012) reported a significant correlation between MTB race times and maximal peak power and mean average power attained in a variant of the Wingate anaerobic test involving five 30-s all-out bouts

interspersed with 30-s of recovery. Similar findings were reported by Hebisz et al. (2017) and Baumann et al. (2012), who also observed significant correlations between anaerobic performance during repeated 20–30 s cycling sprints similar in design to the Wingate test and race times in a simulated competition performed at an intensity above the ventilatory threshold. Zarzeczny et al. (2013) evaluated the maximum power and work output on a single Wingate test among well-trained mountain bikers. They have not observed any correlation with the sports performance. However, while the Wingate test is the most commonly administered test in the assessment of anaerobic power and capacity, its design and short duration may not adequately reflect the specificity of mountain biking. Instead, incremental exercise tests of longer duration may better assess anaerobic capacity in which the intermediate markers of anaerobic glycolysis can be measured including blood hydrogen (H^+) and lactate (La^-) ion concentrations (Desgorces et al. 2007; Juel 2001) or temporal variables associated the plateau at maximal oxygen uptake (Duncan et al. 1996; Gordon et al. 2011).

A review of the literature revealed deficiencies as to the differences in the anaerobic characteristics of high-performance road and mountain bike cyclists, with little data reported on efficiency of the anaerobic energy pathways during high-intensity exercise. Therefore, the purpose of this study was to compare markers of glycolytic metabolism in response to the Wingate test and an incremental test in cohorts with similar maximal oxygen uptake values. It was hypothesized that MTB cyclists, due to specificity of the cycling format, would show increased time of maximal oxygen uptake plateau in incremental test, increased maximal and average power in Wingate test, and higher post-exercise blood La^- and H^+ concentrations in both tests. The basis for this hypothesis is the different characteristics of road and mountain bike racing, in junior categories already. Road racing is longer and less intense in comparison to the mountain bike racing. In turn, mountain bike racing, take place on mountain and forest paths, sometimes with a steep slope, which forces the maximum intensity of effort.

Methods

Participants

Thirty-two male road cyclists (RC) and mountain bike cyclists (MC) were initially recruited. Participants were junior- and youth-category (16–19 years) racers of the National Team. Each had a minimum of 2 years competitive experience in their respective cycling discipline, training on average 8–12 hours per week and competing in 20–25 races per year. Inclusion criteria required approximately 80% of training volume in the 2 months preceding study outset to be performed at an intensity zone below the ventilatory threshold (determined by V-slope analysis of oxygen and carbon dioxide uptake as outlined by Beaver et al. 1986) and maximal oxygen uptake ($\text{VO}_{2\text{max}}$) values between 62–71 $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$ and attainment of plateau at $\text{VO}_{2\text{max}}$ during incremental testing (post-recruitment inclusion). Based on imposition of these criteria, group RC involved 13 participants and group MC 14 participants. Group characteristics are presented in Table 1. The study design was approved by the Ethics Committee of the University School of Physical Education, and conducted in accordance with the Declaration of Helsinki. Participants provided their informed consent following an explanation of testing procedures.

Study design

The Wingate test and the incremental testing protocol (ITP) was administered in laboratory conditions (temperature and humidity controlled) at the Exercise Laboratory of the University School of Physical Education (PN-EN ISO 9001:2001 certified). The Wingate test was the first, the subjects did not perform any training during 48 hours preceding the test. After next 48 hours of passive rest, a incremental test was carried out.

a) Wingate test:

Test was performed on a properly calibrated Excalibur Sport cycle ergometer (Lode B.V., The Netherlands). The test was preceded by a 15-min warm-up with moderate intensity (65% of predicted maximal heart rate). An active 5 minute cool-down was performed by cycling at a low intensity following the warm-up. The Wingate test lasting for 30 s with maximal intensity was then used, during which the maximal and average power (P_{max} and P_{av} , respectively) were measured. The test was performed with an individual fixed-mean crank torque of 0.8 N•m per kg of body mass. Arterialized capillary blood from the fingertip was drawn immediately 3 min after the test in order to assay H^+ with the use of a RapidLab 348 blood gas analyzer (Siemens Healthcare, Germany) and La^- using a Lactate Scout device (SensLab, Leipzig, Germany).

b) Incremental test:

The test was also carried out on the Excalibur Sport cycle ergometer. Starting workload was 50 W and increased every 3 min by 50 W until volitional exhaustion. Time and instantaneous power output were continually recorded via manufacturer software. If the participant was unable to complete a 3-min 50 W stretch, 0.28 W was subtracted for each missing second from the current level of power to determine maximal aerobic power (AP_{max}).

Respiratory function was measured breath-by-breath using a Quark gas analyzer (Cosmed, Italy, Mediolan). The device was calibrated before use with a reference gas mixture of carbon dioxide (5%), oxygen (16%), and nitrogen (79%). Respiratory gas measures were collected 2 min prior test start and continued 3 min after termination and averaged over 30-s intervals. Ventilation was analyzed to determine oxygen uptake (VO_2), carbon dioxide excretion (VCO_2), and minute pulmonary ventilation (VE). Absolute and relative (per kg of body mass) maximal oxygen uptake (VO_{2max}) and maximal carbon dioxide excretion (VCO_{2max}) were calculated based on the composition of expired air and VE . VCO_2 and VO_2

were also used to calculate the respiratory exchange ratio ($RER = VCO_2/VO_2$). Resting values of VO_2 , VCO_2 , and RER were also collected for 3 min post-test (VO_{2post} , VCO_{2post} , and RER_{post} , respectively). The plateau response was determined using previously established methods and designated as the period when VO_2 did not fluctuate $\leq 1.5 \text{ ml.kg}^{-1}.\text{min}^{-1}$ from the VO_{2max} (Doherty et al. 2003; Lucia et al. 2006). This analysis was performed when averaging data every 15 seconds. The time of plateau VO_{2max} incidence ($T_{plateau}$) was calculated in seconds. Additionally, the slope of the logarithmic relationship between VO_2 and work rate was defined until two time points: achievement of P_{max} ($SL_{P_{max}}$) and the onset of the VO_{2max} plateau ($SL_{plateau}$). Similar to the Wingate test, arterialized capillary blood from the fingertip was drawn immediately 3 min after the test in order to determine H^+ with the use of a RapidLab 348 blood gas analyzer and La^- using a Lactate Scout device.

Statistical analysis

Data processing was performed with the Statistica 10.0 software package (Statsoft, Poland). Basic descriptive statistics (means and standard deviations) were calculated for all variables. Evaluation of differences between group means was conducted by using Student's t test. Statistical significance was set at the 0.05 level of confidence in all procedures.

Results

Significant differences between group RC and MC are presented in Table 2 and Figure 1. $T_{plateau}$ was significantly greater in MC than RC ($p = 0.012$) whereas $SL_{P_{max}}$ was significantly lower than RC ($p = 0.01$). Group MC was observed with significantly higher post-ITP values of VO_{2post} ($p = 0.0002$) and VCO_{2post} ($p = 0.009$) and significantly lower RER_{post} ($p = 0.0004$) than group RC. Additionally, post-ITP La^- concentration in MC was

significantly greater than in group RC ($p = 0.046$). The remaining variables -absolute and relative VO_2max , absolute and relative VCO_2max , VE_{max} , RER, AP_{max} , $\text{SL}_{\text{plateau}}$, and H^+ - showed no changes (Table 2, Figure 1).

There were no significant changes in maximal power, average power, and La^- and H^+ concentrations measured during the Wingate test (Table 2).

Discussion

The results of the current study indicate that cross-country MTB cyclists can perform more work than road cyclists at the plateau phase of VO_2max . There are suggestions that plateau incidence can serve as a marker of increased anaerobic substrate metabolism during progressive incremental exercise with no change in VO_2 (Bassett and Howley 2000; Gordon et al. 2015; Lucia et al. 2006). However, data on the temporal characteristics of the VO_2max plateau phenomenon is limited particularly with regard to athlete performance level, sporting discipline, or physiological output characteristics. To date, the majority of the research has focused on the incidence of plateau at VO_2max in trained and untrained individuals (Astorino et al. 2000; Doherty et al. 2003; Gordon et al. 2012). The discussion pertains largely to variance in plateau incidence and reasons explaining lack of attainment. Gordon et al. (2012) believe that the incidence of plateau is dependent on the exercise modality (treadmill vs. cycle ergometry-based incremental testing). An investigation by Astorino et al. (2000) also highlighted the importance of selecting an appropriate sampling interval for the detection of VO_2max plateau.

It has been suggested by Dempsey and Wagner (1999) that athletes with high physiological capacity (e.g. $\text{VO}_2\text{max} > 65 \text{ ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$) are able to attain plateau at VO_2max as these individuals can tolerate the fatigue and pain experienced during such exercise and have the

anaerobic capacity needed in the final phase of ITPs. However, Doherty et al. (2003) reported otherwise in a group of elite middle- and long-distance runners. Even with $\text{VO}_{2\text{max}}$ magnitudes above $65 \text{ ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$, the plateau was identified in only 32% of the participants. Another study by Lucia et al. (2006) found that approximately half of their sample of top-level male professional road cyclists attained $\text{VO}_{2\text{max}}$ plateau. In the present study, from the initial sample of 32 cyclists, five individuals from group RC were excluded from final analysis due to the absence of a $\text{VO}_{2\text{max}}$ plateau. The above data show that not everyone is able to achieve the plateau phase of maximal oxygen uptake, which, according to Dempsey and Wagner (1999), is a confirmation of reaching of the maximal oxygen uptake. Therefore, some believe that in order to confirm the $\text{VO}_{2\text{max}}$ value, the incremental test protocol should be complemented with an additional effort performed after the incremental test. It consists in making several minutes of effort at intensity of about 110% of the work rate achieved on the initial incremental test (Poole and Jones 2017). Similar suggestions result from research by Hill et al. (2002; 2003), where the authors prove that an effort of 3-8 minutes with an intensity of 136% of the maximum power achieved in the incremental test was needed in the examined group to achieve $\text{VO}_{2\text{max}}$ (Hill et al. 2002). In the presented studies, $\text{VO}_{2\text{max}}$ validation was not carried out because the results of only those athletes, who achieved the $\text{VO}_{2\text{max}}$ plateau, were analyzed. However, the $\text{VO}_{2\text{max}}$ plateau phase did not occur in some road cyclists (their results were not analyzed), what confirms the need to verify the achievement of $\text{VO}_{2\text{max}}$ in tests on cyclists.

In comparison to other studies, in the presented studies the much larger proportion of athletes achieved the $\text{VO}_{2\text{max}}$ plateau phase in the incremental test. It is possible it is related to the type of test protocol used. The load in the incremental test used in the present study was increased every 3 minutes, whereas in case of the protocols by Lucia et al. (2006) and Doherty et al. (2003) they increased the power output every 1 minute until volitional

exhaustion. Furthermore, differences in the specificity of road vs. mountain bike cycling may have also influenced incidence and duration of VO_2max plateau regardless of similar training history and performance level. Additional research is needed to understand the relationship between the VO_2max plateau phase and athlete exercise capacity across cycling disciplines to enhance monitoring of training adaptations using incremental testing.

Analysis of the biochemical changes post-ITP revealed that MC had significantly higher La^- than group RC, suggesting greater anaerobic contribution as La^- is a sensitive biomarker of non-oxidative glycolysis (Cairns 2006). Lucia et al. (2006) found that the elite male RC who attained VO_2max plateau also presented higher La^- than those who did not. Reis et al. (2016) reported that the lactate increase by $1 \text{ mmol}\cdot\text{l}^{-1}$ is equal to the energy equivalent of $3 \text{ ml O}_2 \cdot \text{min}^{-1}\cdot\text{kg}^{-1}$. In the presented study, the lactate concentration in the group of mountain bike cyclists was higher on average by $2 \text{ mmol}\cdot\text{l}^{-1}$, so, according to the above report, it may indicate the additional energy production. Moreover, the presented studies showed higher post-ITP $\text{VO}_{2\text{post}}$ and $\text{VCO}_{2\text{post}}$ values in group MC. Based on the classic concept of Hill and Lupton (1923), elevated post-exercise oxygen uptake is indicative of oxygen debt, which is the result of phosphocreatine resynthesis and lactate metabolism processes. However, the concept of oxygen debt has been recently undermined. Currently, it is considered that the metabolic basis of the elevated post-exercise VO_2 may be understood in terms of few factors such as catecholamines, thyroxine, glucocorticoids, fatty acids, calcium ions, and temperature (Gaesser and Brooks 1984). Concomitantly, the lower RER_{post} in group MC (approaching a value of 1) suggests the greater use of La^- as a substrate in oxidative metabolism (Emhoff et al. 2013; Gladden 2008; Robergs et al. 2004) and also improvements in the shuttling of lactate at the intracellular and extracellular level so to be utilized via oxidation or converted to glucose and glycogen (Brooks 2000; Brooks 2007). It is therefore possible that the specificity of cross-country MTB training and competition can

enhance lactate production and oxidization during exercise and improve post-exercise lactate transport capacity from muscle to blood (Impellizzeri et al. 2002; Macdermid and Stannard 2012). Conversely, the longer endurance steady-state activity typical of road cycling could be less conducive to lactate production and efficient disposal. While research does suggest training at a high intensity may stimulate muscle lactate transportation (monocarboxylate transporter MCT1) (Evertsen et al. 2001), other studies have reported that it is long-term continuous endurance training that improves lactate removal by oxidation and gluconeogenesis (MacRae et al. 1995).

Interestingly, while the level of La^- should be proportional to the magnitude of H^+ (Böning and Maassen 2008; Cairns 2006), no significant exercise-induced differences were observed between MC and RC. If La^- was different between MC and RC, the similar H^+ concentrations suggest that MC may present enhanced buffering capacity. One marker of La^- and H^+ clearance is VCO_2 output relative to VO_2 (Peronnet and Aquilaniu 2006; Wasserman et al. 1994; Zhang et al. 1994). However, no significant differences were observed in absolute and relative $\text{VCO}_{2\text{max}}$, while RER_{post} was greater in group RC than MC. Hence, it can be inferred that group MC does not show greater efficacy of buffering by bicarbonate. If this group was able to neutralize H^+ better, it could be probably explained by a non-bicarbonate buffering mechanism. While Gross et al. (2014) and Laursen et al. (2007) did report that high-intensity training induced greater intramuscular buffering capacity than high-volume training following biopsy of exercising muscle, there is no consensus on which intramuscular buffering mechanism is enhanced by this training modality (Sahlin 2014). Derave et al. (2010) credit long-term high-intensity training with increased content of muscle carnosine, an endogenous intracellular buffer, which may explain the differences between RC and MC. Other authors indicate that the increased value of the respiratory exchange ratio is associated with many factors, not only with buffering processes. The amount of carbon dioxide present

in tissues before exercise, the difference in the kinetics between VO_2 and VCO_2 , pulmonary blood flow, and bicarbonate resources are mentioned as those factors (Whipp et al. 1982).

In the presented paper, the markers indicating the involvement of glycolytic anaerobic metabolism, not only during the progressive test, but also in the Wingate test, were sought. Although this is a typical test for anaerobic capacity assessment, no significant differences in measured parameters were found during this test between the examined groups. Some authors recommend the use of the Critical Power (CP) model to assess anaerobic capacity (Jones et al. 2010; Skiba et al. 2014). It was found that CP occurs at an intensity of 80% $\text{VO}_{2\text{max}}$, approximately halfway between the lactate threshold and $\text{VO}_{2\text{max}}$. CP represents the highest power that can be maintained with a constant disturbance of homeostasis, i.e. 1) no further decrease phosphocreatine, muscle and blood pH, and bicarbonates; 2) no further increase in the lactate in blood, oxygen uptake, and minute pulmonary ventilation (Jones et al., 2010). The determination of CP may be helpful in assessing the capacity level; therefore, determining CP in the group of road and mountain bike cyclists will be additional aim when planning the next tests, to observe if this indicator can differentiate the examined groups.

This study demonstrated that the differences between the modalities of road and mountain bike cycling are reflected in the anaerobic characteristics of athletes assessed during an incremental exercise test. The application of incremental testing shows validity in the measurement of anaerobic capacity concomitant to aerobic capacity. Especially when no significant changes in the Wingate test are observed. The obtained data may be useful to coaches and competitive athletes who monitor training adaptations via incremental testing.

We concluded that, compared with road cyclists, mountain bike cyclists present increased time of $\text{VO}_{2\text{max}}$ plateau, greater post-exercise blood lactate concentration, and higher post-exercise oxygen uptake. These results suggest greater anaerobic contribution during incremental testing efforts.

Conflict of Interest

There is no conflict of interest.

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Table 1. Anthropometric and physiological characteristics of RC and MC

Group	Age [years]	Body height [cm]	Body mass [kg]	VO ₂ max [ml·min ⁻¹ ·kg ⁻¹]	Pmax [W]
RC	17.8 ± 0.6	180.4 ± 4.1	69.2 ± 4.9	65.5 ± 2.3	378.9 ± 25
MC	17.9 ± 1.2	178.2 ± 4.1	68.8 ± 6	66.3 ± 2.5	390 ± 36.9

VO₂max – maximal oxygen uptake determined during incremental testing protocol, Pmax – maximal power output determined during incremental testing protocol, RC – road cyclists, MC – mountain bike cyclists, data are presented as mean ± standard deviation

Table 2. Differences between road cyclists and mountain bike cyclists in physiological- and performance-derived responses to Incremental testing protocol and Wingate test

Variable	Road cyclists	Mountain-bike cyclists
Incremental testing protocol		
SL _{Pmax} [ml·min ⁻¹ ·W ⁻¹]	10.88 ± 0.43	10.3 ± 0.64*
VO _{2max} [l·min ⁻¹]	4.53 ± 0.36	4.56 ± 0.41
VO _{2post} [l·3min ⁻¹]	4.63 ± 0.52	5.74 ± 0.77**
SL _{plateau} [ml·min ⁻¹ ·W ⁻¹]	11.28 ± 0.53	10.96 ± 0.45
VCO _{2max} [l·min ⁻¹]	5.31 ± 0.4	5.22 ± 0.51
VCO _{2max} [ml·min ⁻¹ ·kg ⁻¹]	76.7 ± 3.4	75.8 ± 3.8
VCO _{2max} [l·3min ⁻¹]	15.1 ± 1.24	15.1 ± 1.42
VCO _{2post} [l·3min ⁻¹]	6.75 ± 0.65	7.8 ± 1.2**
RER	1.17 ± 0.03	1.14 ± 0.05
RERpost	1.46 ± 0.06	1.36 ± 0.06**
VE _{max} [l·min ⁻¹]	162.8 ± 16.3	174.7 ± 29.3
H ⁺ [nmol·l ⁻¹]	63.5 ± 4.27	66.7 ± 8.0
Wingate test		
P _{max} [W]	1407.48 ± 140.69	1332.41 ± 203.48
P _{max} [W·kg ⁻¹]	20.33 ± 0.96	19.36 ± 2.68
P _{av} [W]	751.35 ± 70.31	745.61 ± 83.77
P _{av} [W·kg ⁻¹]	10.85 ± 0.42	10.83 ± 0.86
La ⁻ [mmol·l ⁻¹]	15.68 ± 1.8	14.98 ± 2.8
H ⁺ [nmol·l ⁻¹]	73.26 ± 4.8	68.36 ± 6.7

SL_{Pmax} - the slope of relationship between oxygen uptake and work rate, until achievement of maximal aerobic power, VO_{2max} – maximal oxygen uptake, VO_{2post} – post-exercise oxygen uptake for 3 min, SL_{plateau} - the slope of relationship between oxygen uptake and work rate, until achievement of VO_{2max} plateau, VCO_{2max} – maximal carbon dioxide excretion, VCO_{2post} – post-exercise carbon dioxide excretion for 3 min, RER – respiratory exchange ratio, RERpost – post-exercise respiratory exchange ratio, VE_{max} – maximal minute pulmonary ventilation, La⁻ - lactate ion concentration, H⁺ - hydrogen ion concentration, P_{max} – maximal power, P_{av} – average power, * – significant difference between RC vs. MC at $p < 0.05$, ** – significant difference between RC vs. MC at $p < 0.01$; data are means ± standard deviations

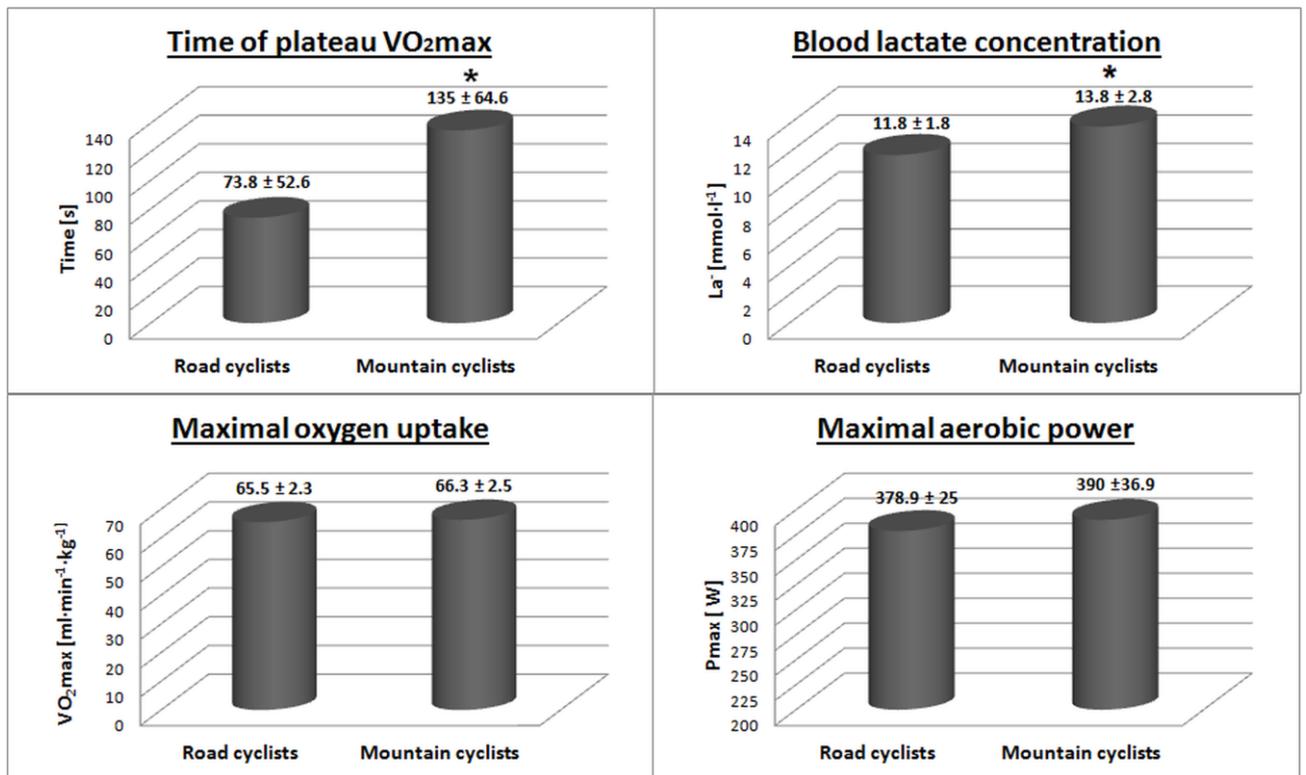


Figure 1. Differences between road cyclists and mountain bike cyclists in selected parameters responses to Incremental testing protocol