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Comparison of physiological responses and muscle activity during incremental and decremental cycling exercise

Submission type: original investigation

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Running title: Decremental exercise tests in cyclists

Abstract word count: 224
Text-only Word Count: 2949
Number of figures and tables: 7

Funding Disclosure: This work was funded by the funded by Discovery Health (Pty) (Ltd), the Medical Research Council of South Africa, the Harry Crossley and Nellie Atkinson Staff Research Funds of the University of Cape Town, and the National Research Foundation of South Africa through its THRIP initiative.

Abstract

Objective: to investigate whether a cycling test based on decremental loads (DEC) could elicit higher $\dot{V}O_{2max}$ values compared with an incremental test (INC).

Design: Nineteen well-trained individuals performed an INC and a DEC test on a single day, in randomized order.

Methods: During INC the load was increased by 20 $W.min^{-1}$ until task failure. During DEC the load started at 20 W higher than the peak load achieved during INC (familiarization trial) and was progressively decreased. Gas exchange and EMG activity (n=11) from four lower limb muscles were monitored throughout the tests. Physiological and EMG data measured at $\dot{V}O_{2max}$ were compared between the two protocols using paired t-tests.

Results: $\dot{V}O_{2max}$ during the DEC was 3.0 (5.9) % higher than during INC (range 94 – 116 %; $p = 0.01$), in spite of a lower power output (-21 [20] W, $p < 0.001$) at $\dot{V}O_{2max}$. Pulmonary ventilation ($p = 0.036$) and breathing rate ($p = 0.023$) were also higher during DEC. EMG activity measured at $\dot{V}O_{2max}$ was not different between tests, despite the lower output during DEC.

Conclusions: A decremental exercise test produces higher $\dot{V}O_{2max}$ in cycling compared to an incremental test, which was accompanied by higher pulmonary ventilation and similar EMG activity. The additional O_2 uptake during DEC might be related to extra work performed by the respiratory and the less oxidatively efficient leg muscles.

Key-words: maximal oxygen uptake, $\dot{V}O_{2max}$ plateau, exercise testing, decremental, cycling

Introduction

Maximal oxygen uptake ($\dot{V}O_{2max}$) represents the upper limit of cardiorespiratory capacity during exercise. Because of its association with several performance and even health indicators, it is important that $\dot{V}O_{2max}$ is properly assessed during testing, and a substantial body of literature has been dedicated to it [1]. Current standards dictate that $\dot{V}O_{2max}$ should be assessed during incremental exercise tests, in which the workload increases at given time intervals until the participant is exhausted. In these tests, leveling-off of $\dot{V}O_2$ with increasing exercise intensities – the $\dot{V}O_{2max}$ plateau phenomenon – is seen as evidence that $\dot{V}O_{2max}$ is, indeed, maximal. When evidence of a plateau is not present, a second test at higher workload is recommended to confirm the attainment of $\dot{V}O_{2max}$, the so-called verification phase or verification test [1].

A few studies, however, have shown that alternatives to the incremental exercise test can produce even higher values of $\dot{V}O_{2max}$. Some of these tests are based on cycling time-trial efforts [2,3] or self-paced and closed-loop exercise [4], and even decremental exercise tests in runners [5] and elite runners [6]. While these studies have called into question whether the $\dot{V}O_{2max}$ measured during incremental tests (with or without the verification phase) truly represents a cardiorespiratory limitation, later studies have not always corroborated these findings, for instance showing no difference between traditional incremental tests and self-paced tests in cycling [7,8] or decremental tests in runners [9].

Some these contradicting results might be specific to the conditions of each particular study. For example, Jenkins and colleagues found that a self-paced protocol produced higher $\dot{V}O_{2max}$ values in younger, but not in older individuals [10]. Furthermore, $\dot{V}O_{2max}$ not only varies according to one's training background [11], but is also specific to a given exercise modality [8]. In this context, to date only one independent attempt has been made to replicate the finding of a higher $\dot{V}O_{2max}$ during a decremental test, also in runners [9]. Therefore, the aim of this investigation was to test whether the findings of a higher $\dot{V}O_{2max}$ during decremental running exercise tests originally reported in well-trained individuals [5] would also be present in a group of well-trained cyclists. In addition, we wished to determine whether muscle activity of the lower limbs would be different at the time that $\dot{V}O_{2max}$ was achieved in both tests. We postulated that a higher $\dot{V}O_{2max}$ would be present during the decremental exercise test and that in spite of a lower power output at $\dot{V}O_{2max}$ the EMG activity would be higher, indicating a greater muscle activation.

Methods

Subjects

The study was approved by the Ethics and Research Committee of the Faculty of Health Sciences of the University of Cape Town (253/2009). Participants were clearly informed about the risks and procedures involved in this investigation and gave their written consent for participation. The study was divided in two parts (see figure 1). In part A, eleven participants (ten male and one female, (mean [SD]) age 29.0 [7.4] years, body mass 75.2 [7.8] kg, height 181 [9] cm), all accustomed to high-intensity cycling, visited the laboratory on two occasions. In part B, eight male participants (age 23.8 [5.5] years, body mass 69.8 [8.1] kg, height 179 [4] cm), all accustomed to high-intensity cycling, visited the laboratory on three occasions, each separated by 2-3 days.

Design

This investigation was set-up as repeated measures design where $\dot{V}O_{2max}$ was defined as the dependent variable and primary outcome and the exercise test (i.e. incremental or decremental exercise test) as the independent variable. Both exercise tests were performed in the same day (see below) and order in which the tests were performed was randomized and balanced between participants, so that an equal number of participants performed each of the two test orders. Other outcomes of interest were gas exchange, heart rate and workload at the time of $\dot{V}O_{2max}$.

Furthermore, based on prior data from our group that showed that both trained [5] and elite runners [6] showed a higher $\dot{V}O_{2max}$ during an incremental test following two exposures to the decremental test (in contrast to a control group with stable $\dot{V}O_{2max}$ over five consecutive incremental test trials), we sub-divided the present study into two parts (Figure 1). Part A was designed to ensure no adaptative effect from the decremental test could take place (i.e. no familiarization with the protocol was allowed prior to the main comparison), whereas in Part B an additional visit was included where participants were familiarized with the decremental test.

In part A, the participants visited the laboratory on two occasions. On their first visit (familiarization), the participants performed a maximal incremental exercise test on a cycle ergometer (INC_{fam}), followed 15 minutes later by a verification test to confirm the attainment of $\dot{V}O_{2max}$. On visit two, the participants performed three exercise tests, each separated by approximately one hour. First, they performed an incremental test (INC_1), following which a decremental (DEC) and another incremental test (INC) were performed in randomized, counter-balanced order. During the trials on visit two, gas exchange data, heart rate and EMG activity of four leg muscles (*vastus lateralis*, *biceps femoris*, v*astus medialis* and *gastrocnemius medialis*) were continuously monitored. Prior to the first incremental test, a 10-s sprint was performed, with the ergometer set in linear (cadence-dependent) mode. This sprint served as a reference for the EMG analysis in the time domain, as it has been recommended [12].

In part B, the participants visited the laboratory on three occasions, each separated by 2-3 days. The first visit was similar to that described in part A, and on the second visit a familiarization with the decremental exercise test (DEC_{fam}) was performed. On their third visit, the participants repeated the INC and DEC exercise tests in randomized, counter-balanced order, separated by at least 60 minutes of rest. Gas exchange data, heart rate and power output were monitored throughout the tests.

Cycling tests

All exercise trials were performed on an electronically braked cycle ergometer (Lode Excalibur Sport, Lode, Netherlands). Prior to each exercise test, the participants warmed-up for 10 minutes at a self-selected cadence and power output. The INC tests started at 200 W for men and 160 W for women, with increments of 20 W applied every 60 seconds thereafter, until cadence fell below 60 revolutions per minute (rpm). Strong verbal encouragement was given whenever the pedaling cadence approached the 60-rpm mark. The verification test consisted of a square-wave test to exhaustion performed at one stage higher (20 W) than the last completed stage during INC_{fam}. Immediately prior to the test, the participants cycled for three minutes at 150 W (120 W for women). The load was then increased to the desired power within 20 seconds, and participants were instructed to cycle for as long as they could. The DEC test started with three minutes of light cycling (150 W for men, 120 W for women), following which the load was raised within 20 seconds to the equivalent used during the verification test (that is, 20 W higher than the load of the last completed stage during INC_{fam}). The initial load was maintained for 60%

of each individual's time to task failure recorded during the verification phase test. The average duration for the first stage of DEC was 90 s (range 75-105 s). Following the first stage, the load was decreased by 20 W and maintained for 30 s. Subsequent decrements of 10 W were maintained for 45, 60, 90 and 120 s, thus totaling ~7 min 15 s of test time. If this protocol proved too demanding for the subject (time to fatigue < 3 minutes during DEC_{fam}, part B only), on the second occasion that they performed the decremental protocol (DEC, Part B) the first decrement in workload was increased from 20 to 30 W.

Cardiorespiratory variables

Gas exchange and heart rate were continuously monitored during the exercise tests using calibrated metabolic carts. In Part A, a MOXUS Modular Metabolic System was used (AEI Technologies, IL, USA) paired with a Polar S410 (Polar Electro OY, Kempele, Finland). In Part B, a Cortex Metalyser was used (3B, Biophysik, Leipzig, Germany) paired with a Polar T31. All gas exchange and heart rate data exported as consecutive 5 s means. $\dot{V}O_{2max}$ was defined as the highest 60 s moving average measured during a test and all other physiological variables were recorded at the same time as $\dot{V}O_{2max}$. For comparison of $\dot{V}O_{2max}$ values on day 1, between the incremental and verification tests, 30-s moving averages were used in order to compensate for the much shorter duration of the verification tests, where $\dot{V}O_{2max}$ was not necessarily constant for a full minute.

Muscle electrical activity

In Part A, EMG signals from the *vastus lateralis*, *vastus medialis*, *gastrocnemius medialis* and *biceps femoris* of the right leg were continuously recorded (Part A) using DE-2.1 single differential surface sensors with 10 mm inter-electrode distance (Delsys Inc., Boston, MA, USA). SENIAM [13] and Delsys recommendations for the placement of the sensors on the skin were followed. Before electrode placement, the skin was shaved to remove excessive hair and wiped with isopropyl alcohol. A reference electrode was applied over the patella. The EMG signals were sampled at 2000 Hz and amplified using Bagnoli-8 (Delsys Inc). The EMG signals were transferred into Power Lab (ADInstruments, Colorado Springs, CO, US) and filtered using a 15-500 Hz band pass filter in Lab Chart Pro software (ADInstruments). Root mean square EMG activity was calculated for every 20 ms and averaged over the minute in which $\dot{V}O_{2max}$ was measured. Only the periods of active contraction were used for analysis, which were determined as root mean square EMG values > 10% above baseline (quiet sitting on ergometer). EMG activity was expressed as a percentage of the root mean square activity recorded during the 10-s sprint which was performed immediately prior to the first exercise trial (but after the 10 min warm-up period). The % activation was also multiplied by the fraction of time in which the muscle was active to obtain an estimate of total activation, akin to the integrated EMG signal [14]. The frequency contents of the EMG signal were analyzed using fast Fourier transformation, and the mean and median frequency from the EMG signal during the period in which $\dot{V}O_{2max}$ was measured were determined. Prior to the sprint, the participants cycled at a low power output for two minutes. EMG data is presented as n = 7 for the *vastus lateralis*, *gastrocnemius medialis* and *biceps femoris* and n = 9 for the *vastus medialis* due to technically inadequate data for at least one test on some participants, usually the result of electrode detachment/excessive noise mid-test.

Statistics

All data are presented as mean (standard deviation), unless stated otherwise. The reproducibility of $\dot{V}O_{2max}$ and EMG variables between INC_1 (performed before the experimental trials) and INC (performed in randomized order with DEC) was analyzed by calculating the intraclass correlation coefficient (ICC), the typical error of

measurement (the standard deviation of the difference between measurements divided by the square root of two) and the individual coefficient of variation [15]. The physiological responses from the participants in both parts of the study were pooled together and the physiological responses between the DEC and INC analyzed used Student's paired t-tests, with the normality of residuals analyzed using Q-Q plots. Comparisons are presented with the t-statistic, 95% confidence interval of the differences (95% CI) and the effect size as Cohen's d for independent samples (mean difference divided by the pooled standard deviation of both groups) or Cohen d_z for dependent samples (mean of differences divided by the standard deviation of the difference scores). The effect of adding a specific familiarization trial for the decremental test (part B vs. part A) was analyzed by comparing the DEC/INC $\dot{V}O_{2max}$ ratio in parts A and B of the study, using Student's independent t-test. Significance level was set at $p < 0.05$, and all statistical procedures were calculated using Prism 8.3 (GraphPad, La Jolla, CA) or dedicated Excel spreadsheets.

Results

$\dot{V}O_{2max}$ *during the incremental and verification tests*

During INC, participants were able to exercise for 8.3 (1.7) minutes reaching a $\dot{V}O_{2max}$ of 55.8 (8.2) $mL.kg^{-1}.min^{-1}$(parts A and B pooled). In the familiarization visits, $\dot{V}O_{2max}$ obtained from the incremental test was not different from that measured during the verification tests (30-s moving averages, 4,139 [743] vs. 3,989 [717] $mL.min^{-1}$ respectively, 95% CI -345 – 46 $mL.min^{-1}$, p = 0.125). 30-s moving averages of $\dot{V}O_2$were used in both tests due to the much shorter nature of the verification test. When the $\dot{V}O_{2max}$ of the familiarization session as compared with that from the main experimental session, there was no statistical difference between the two (mean difference 0.0 $mL.min^{-1}$, 95% CI -195 – 196 $mL.min^{-1}$, p = 0.99). This is in spite of a significant difference increase in exercise tolerance on the second attempt (+ 0.6 min, 95% CI 0.2 – 1.0 min, p = 0.008).

On the experimental day of part A of the study, when two incremental tests were performed (INC_1 and INC), $\dot{V}O_{2max}$ values were very similar between them (4,366 [464] vs. 4,354 [499] $mL.min^{-1}$ for INC_1 and INC respectively, 95% CI -80 – 57 $mL.min^{-1}$, p = 0.716). Intra-class coefficient for $\dot{V}O_{2max}$ (INC_1 vs INC) was 0.98, and the typical error of measurement was 72 $mL.min^{-1}$ (1.7%).

$\dot{V}O_{2max}$ during the decremental test

During DEC (parts A and B pooled), $\dot{V}O_{2max}$ values were on average 3.0% higher than during INC (4,259 [695] vs. 4,139 [657] $mL.min^{-1}$ respectively, 95% CI 16 – 224 $ml.min^{-}1$, $d_z = 0.56$, p = 0.025, Figure 2), although they were exercising at a significantly lower power output at the time that $\dot{V}O_{2max}$ was achieved (312 [43] vs. 333 [47] W for DEC and INC respectively, 95% CI 10.8 – 31.1 W, $d_z = 1.03$, $p < 0.001$). Individual data for 60-s moving averages in both tests are provided in supplemental figure 1. An additional comparison was performed between DEC and INC using 30-s moving averages and yielded similar results (data not shown). Test termination during DEC occurred at 5.7 (1.6) min, (range 2.5 – 9.5 min, not considering the preceding 3 min lead-in pahse at moderate power) which was significantly shorter than the INC test (-2.9 min, 95% CI -4.3 – -1.6 min, p = 0.0002). Important, two decremental tests were interrupted by the experimenter as participants reached the end of the protocol. All physiological data measured during the exercise tests are shown in table 2. During DEC, participants also reached 4.5% higher pulmonary ventilation (V_E) in comparison to INC (p = 0.036), due

to an increased breathing rate (BR) (Table 2, p = 0.023). Respiratory exchange ratio, carbon dioxide output and heart rate were not different between the two exercise tests at $\dot{V}O_{2max}$ (Table 2).

Effects of familiarization on the decremental test

In Part B, there were no differences in $\dot{V}O_{2max}$ between DEC_{fam} and DEC (3,904 [718] vs. 3,953 [758] $mL.min^{-1}$ respectively, 95% CI -17 – 71 $ml.min^{-1}$, $d_z = 0.34$, n = 8, p = 0.371). Likewise, when parts A and B were compared with regards to the ratio of DEC/INC $\dot{V}O_{2max}$ elicited there were no significant differences (1.017 [0.042] vs. 1.047 [0.069] for parts A and B respectively, 95% CI -0.02 – 0.08, d = 0.54, p = 0.258).

Muscle electrical activity

For the EMG variables, reliability data for each muscle between INC_1 and INC are shown in Table 1. Overall, all four analyzed muscles showed a good degree of reliability (ICC 0.85 – 0.98).

Individual data for EMG activity of the four evaluated muscles is presented in Figure 3. There were no significant differences in mean EMG activity or total activation measured at $\dot{V}O_{2max}$ between INC and DEC for any of the analyzed muscle groups. There were also no significant differences between INC and DEC for any of the muscles for mean or median frequency (Table 3).

Discussion

The main findings from this study are that: i) trained subjects achieved higher $\dot{V}O_{2max}$ values during a cycling DEC in comparison to INC; ii) the higher $\dot{V}O_{2max}$ during DEC was accompanied by higher $\dot{V}_E$ in comparison to INC; iii) EMG activity for four lower limb muscles was similar at the time of $\dot{V}O_{2max}$ in both testing protocols; and iv) the order of the tests or previous familiarization with DEC did not affect the $\dot{V}O_{2max}$ results.

The ideal protocol for the assessment $\dot{V}O_{2max}$ has been the topic of research for years. Under the current paradigm of incremental tests, it has become clear that $\dot{V}O_{2max}$ is robust over a wide range of protocols and test durations [16]. Furthermore, it has also been shown that simply requiring higher energetic output does not result in higher $\dot{V}O_{2max}$, whether the additional output comes from the exercising limbs, as in the case of the verification test [17], or the upper arms [18]. Our data confirms these findings, as the values obtained during the verification tests were if anything lower (although the difference did not reach statistical significance) than those measured during the preceding incremental test. Thus, the $\dot{V}O_{2max}$ determined during the incremental test in our study is within what is currently considered best practice in terms of protocol and analysis.

In this context, the decremental exercise actually operates within a similar range of workloads, as the test starts at the same workload as the verification test and only goes down. This eliminates the possibility that any differences between protocols would the result of a poor choice of work rate increase from the incremental tests. Based on our previous research and pilot testing, however, it became evident that a perfect mirror-image of an incremental test would lead to a too rapid decrement in workload, and eventually all cardiorespiratory responses would turn downwards before $\dot{V}O_{2max}$ could be

reached. Exercise intensities within the so-called "severe" domain are known to eventually lead to $\dot{V}O_{2max}$ [19], and in trying to balance a prolongation of the protocol towards more than 2-3 min with eliciting the highest possible physiological response a protocol with decreasing workloads of increased durations was envisioned. Although the decremental and incremental protocol differ in terms of time taken to reach $\dot{V}O_{2max}$ as well as work performed, , this should not prevent a comparison between the two, much like different incremental tests with slightly different ramps are routinely compared and also do not show differences in $\dot{V}O_{2max}$ [16], or even the comparison between all-out exercise and incremental tests [20].

In spite of the lower power output at $\dot{V}O_{2max}$ during DEC, EMG activity was similar between DEC and INC at the time of $\dot{V}O_{2max}$. The increase in $\dot{V}O_2$ during all-out exercise tests (where $\dot{V}O_2$ increases up to $\dot{V}O_{2max}$ despite decreasing power output and EMG activity) has been proposed to be a "slow-component"-like phenomenon [21], resulting from the loss of efficiency in the exercising muscles. $\dot{V}O_2$ then increases during exercise because of progressive recruitment of less efficient, type IIx fiber, which increases the O_2 cost for force production per fiber. An alternative explanation is that while an initial bout of very intense exercise creates the demand for O_2, a subsequent decrease in power output might improve limb blood flow, as muscle contraction intensity and vasoconstriction would be likely reduced, thereby improving O_2 extraction. The similar EMG activity coupled with lower power output found in the present study likely facilitated higher blood flow, which could indeed have facilitated higher O_2 extraction.

To test whether our EMG measures were sensitive enough to detect differences caused by a 20 W change in power output (i.e. the difference between INC and DEC at $\dot{V}O_{2max}$), we compared the root mean square and total activation of the analyzed muscles prior to $\dot{V}O_{2max}$ during the incremental test, since our incremental protocol had a load rate of 20 $W.min^{-1}$. For both the *vastus lateralis* and *vastus medialis*, root mean square and/or total activation were sufficiently sensitive to have detected changes equivalent to a 20 W change in power output, at least during a ramp-up protocol (Table 4). We did not find differences in the mean or median frequency of the EMG signal of any of the analyzed muscles, which precludes any suggestion of change in recruitment pattern. Furthermore, the total EMG activity was similar between INC and DEC, suggesting indeed lower efficiency of the muscle contraction rather than exclusively higher oxidative metabolism *per se*. Although our results suggest a loss of efficiency in producing mechanical work, as shown by a higher $\dot{V}O_2$ for a similar EMG activity, it is not possible to argue conclusively that the measured difference in $\dot{V}O_2$ (~125 $mL.min^{-1}$) is related to changes in muscle efficiency.

The results from the present study extend the findings originally reported during uphill running [5], showing ~3% higher $\dot{V}O_{2max}$ during DEC in comparison to INC when exercise is performed on a cycle ergometer. These results were seen even though participants had performed a verification test, current seen as the best standard to ensure the attainment of "true" maximal oxygen uptake. The additional O_2 uptake requires either additional blood supply and/or increased oxygen extraction to take place during exercise, be it in the respiratory muscles, exercising muscles or elsewhere. Mauger et al. [4] hypothesized that a better distribution of blood flow in the active limbs due to a decrease in power output [22,23]could explain the higher $\dot{V}O_{2max}$ measured during a self-paced test, but this proposition remains purely speculative. In muscle preparations $\dot{V}O_{2max}$ can be altered by increasing O_2 extraction while O_2 delivery is kept constant when independently manipulating SaO_2 and PaO_2, although to our knowledge this not been demonstrated in vivo [24].

In the present study, both $\dot{V}_E$ and BR were higher during DEC at the time of $\dot{V}O_{2max}$, different to the findings of Beltrami [5] and Taylor [9]. Thus, at least in cycling part of the difference in $\dot{V}O_{2max}$ measured between tests might be related to the additional O_2 cost of breathing. The 7.0 L.min^{-1} difference in $\dot{V}_E$ measured between DEC and INC could raise $\dot{V}O_2$ by approximately 20-30 mL.min^{-1} [25], or explain 15-25% of the difference in $\dot{V}O_{2max}$ measured between DEC and INC (120 mL.min^{-1}). It must be noted, however, that differences in $\dot{V}_E$ do not always result in measurable differences in pulmonary $\dot{V}O_2$ [26]. Moreover, simply forcing the respiratory muscles to perform more work does not increase whole-body $\dot{V}O_{2max}$ or cardiac output [27], similar to what happens when an additional (supra-maximal) load is imposed to the muscles of the exercising limbs [18]. Therefore, while the respiratory muscles can be seen as the receiver of the additional $\dot{V}O_2$, it does not serve a mechanistical explanation of how $\dot{V}O_2$ was increased.

The differences in $\dot{V}O_{2max}$ between DEC and INC in the present study were independent of whether participants were allowed a specific familiarization trial for DEC or not and were also independent of the order in which INC and DEC were performed. This contrasts with previous findings, in which following DEC participants retained the higher $\dot{V}O_{2max}$ on a subsequent INC despite an unchanged performance in the final incremental test [5]. Should the same had happened in the present study, $\dot{V}O_{2max}$ would have increased from INC_{fam} to INC (Part B), since participants performed the DEC_{fam} trial between these two tests. Also, if participants had retained the higher $\dot{V}O_{2max}$ values following DEC, the $\dot{V}O_{2max}$ differences between INC and DEC would have been blunted for all participants who performed INC after DEC, which did not happen. Although these findings do not help to explain why the running studies showed a persistent increased $\dot{V}O_{2max}$ following the DEC protocol, they show that future research using cycling exercise can be performed using a familiarization trial for both INC and DEC.

This investigation is not without its limitations. First, the decremental test per se could be criticized for the tailored approach and the duration of the first stage (60% of the time to exhaustion during the verification test). From our experience, this proved to be a good compromise between sufficient intensity to drive O_2 uptake upwards and preventing premature exhaustion. In contrast to our running experiments, anecdotally the cyclists often reported not even feeling the first two (20 W each) drops in workload, but merely a non-worsening of the associated pain and discomfort. While it is entirely possible that a different duration of the first stage – or indeed a different rate of load decrement – would have led to different results, this remains speculative. Another possibility would be the use of a V-shaped protocol as recently proposed [6], however it remains to be seen how tolerable such protocol would be in cycling. Second, participants in part A performed an additional incremental test (always first of the day). The reason for this test was that as participants were unfamiliarized with the decremental protocol, the initial incremental test provided an updated value of peak power output, from which the starting load of the decremental protocol could be more acurately calculated. Although this additional test might have caused some level of fatigue (despite over 60 min of recovery), we note that $\dot{V}O_{2max}$ from this test was not different from the one performed in randomized order with the decremental test. Furthermore, it has been demonstrated that trained individuals can perform up to four incremental tests in a day (with 1.5h passive rest) without losses to $\dot{V}O_{2max}$ or peak power output [28]. Finally, even if some fatigue was present, this would have affected both the decremental and incremental tests, as the order of the two was balanced. At present we see no reason to believe that increased fatigue should lead to higher $\dot{V}O_{2max}$, as if this was the case the verification tests – performed within minutes of an incremental test – should have shown this effect. Lastly, while the decremental

protocol might be able to elicit higher $\dot{V}O_{2max}$ values, an important pitfall of the protocol is that it does not allow for the determination of commonly assessed variables such as ventilatory or lactate thresholds or cycling efficiency, or even peak power output, as this is a function of the starting load and thus imposed by the experimenter.

Practical Applications

Decremental exercise tests open a new paradigm for exercise testing, possibly leading to higher $\dot{V}O_{2max}$ than incremental tests. Muscle activity at $\dot{V}O_{2max}$ suggests that, during cycling tests, decremental protocols lead to greater physiological strain, with loss of metabolic efficiency. While a more optimal protocol might emerge, at present a successful alternative is a starting load equal to VER with 60% of time to exhaustion and two subsequent decreases of 20 W within 30 s in order to prevent exhaustion, after which decreases can be attenuated and stages elongated. It is possible that the $\dot{V}O_{2max}$ measured during a decremental exercise tests presents a different relationship with markers of health and performance, but this remains to be established. Due to the nature of the workload and physiological response, however, the decremental test paradigm is unable to produce other markers that could be of interest in athletic settings, such as lactate or ventilatory thresholds.

Conclusion

Trained individuals achieved higher $\dot{V}O_{2max}$ during a decremental cycling protocol in comparison to a traditional incremental exercise test. The additional $\dot{V}O_2$ may partly be attributed to the extra work performed by the respiratory muscles and less efficient lower limbs, but it remains unclear whether it originates from higher O_2 delivery and/or extraction. These results argue against the notion that $\dot{V}O_{2max}$ as determined from an incremental test represent an absolute ceiling in cardiorespiratory capacity.

Acknowledgements

The authors are grateful to all participants for volunteering their time to this investigation. No specific financial support was used in this investigation.

References

1. Poole DC, Jones AM. CORP: Measurement of the Maximum Oxygen Uptake (VO2max): VO2peak is no longer acceptable. *J Appl Physiol*. 2017:jap.01063.2016. doi:10.1152/japplphysiol.01063.2016

2. Foster C, Green MA, Snyder AC, Thompson NN. Physiological responses during simulated competition. *Med Sci Sports Exerc*. 1993;25(7):877-882. http://europepmc.org/abstract/med/8350712. Accessed August 7, 2014.

3. McGawley K, Holmberg H-C. Aerobic and anaerobic contributions to energy production among junior male and female cross-country skiers during diagonal skiing. *Int J Sports Physiol Perform*. 2014;9(1):32-40. doi:10.1123/ijspp.2013-0239

4. Mauger AR, Metcalfe AJ, Taylor L, Castle PC. The efficacy of the self-paced V̇O2max test to measure maximal oxygen uptake in treadmill running. *Appl Physiol Nutr Metab*. 2013;38(12):1211-1216. doi:10.1139/apnm-2012-0384

5. Beltrami FG, Froyd C, Mauger AR, Metcalfe AJ, Marino F, Noakes TD. Conventional testing methods produce submaximal values of maximum oxygen consumption. *Br J Sports Med*. 2012;46(1):23-29. doi:10.1136/bjsports-2011-090306

6. Beltrami FG, Noakes TD. Conventional Testing Produces Submaximal Values for Oxygen Uptake in Elite Runners. *Int J Sports Physiol Perform*. 2021:1-6. doi:10.1123/ijspp.2020-0778

7. Hogg JS, Hopker JG, Mauger AR. The self-paced VO2max test to assess maximal oxygen uptake in highly trained runners. *Int J Sports Physiol Perform*. 2015;10(2):172-177. doi:10.1123/ijspp.2014-0041

8. Hanson NJ, Scheadler CM, Lee TL, Neuenfeldt NC, Michael TJ, Miller MG. Modality determines VO2max achieved in self-paced exercise tests: validation with the Bruce protocol. *Eur J Appl Physiol*. 2016;116(7):1313-1319. doi:10.1007/s00421-016-3384-0

9. Taylor K, Seegmiller J, Vella CA. The Decremental Protocol as an Alternative Protocol to Measure Maximal Oxygen Consumption in Athletes. *Int J Sports Physiol Perform*. 2016;11(8):1094-1099. doi:10.1123/ijspp.2015-0488

10. Jenkins LA, Mauger AR, Hopker JG. Age differences in physiological responses to self-paced and incremental V˙ O 2 max testing. *Eur J Appl Physiol*. 2017;117(1):159-170. doi:10.1007/s00421-016-3508-6

11. Millet GP, Vleck VE, Bentley DJ. Physiological differences between cycling and running: lessons from triathletes. *Sports Med*. 2009;39(3):179-206. http://www.ncbi.nlm.nih.gov/pubmed/19290675.

12. Jacobs R a, Rasmussen P, Siebenmann C, et al. Determinants of time trial performance and maximal incremental exercise in highly trained endurance athletes. *J Appl Physiol*. 2011;111(5):1422-1430. doi:10.1152/japplphysiol.00625.2011

13. Hermens HJ, Freriks B, Disselhorst-Klug C, Rau G. Development of recommendations for SEMG sensors and sensor placement procedures. *J Electromyogr Kinesiol*. 2000;10(5):361-374. doi:10.1016/S1050-6411(00)00027-4

14. Torres-Peralta R, Morales-Alamo D, González-Izal M, et al. Task Failure during Exercise to Exhaustion in Normoxia and Hypoxia Is Due to Reduced Muscle Activation Caused by Central Mechanisms While Muscle Metaboreflex Does Not Limit Performance. *Front Physiol*. 2016;6(January):1-15. doi:10.3389/fphys.2015.00414

15. Hopkins WG. Measures of reliability in sports medicine and science. *Sports Med*. 2000;30(1):1-15. doi:10.2165/00007256-200030050-00006

16. Zhang YY, Johnson MC, Chow N, Wasserman K. Effect of exercise testing protocol on parameters of aerobic function. *Med Sci Sports Exerc*. 1991;23(5):625-630.

17. Midgley AW, McNaughton LR, Polman R, Marchant D. Criteria for determination of maximal oxygen uptake: a brief critique and recommendations for future research. *Sports Med*. 2007;37(12):1019-1028. http://www.ncbi.nlm.nih.gov/pubmed/18027991.

18. Brink-Elfegoun T, Holmberg H-C, Ekblom MN, Ekblom B. Neuromuscular and circulatory adaptation during combined arm and leg exercise with different maximal work loads. *Eur J Appl Physiol*. 2007;101(5):603-611. doi:10.1007/s00421-007-0526-4

19. Black MI, Jones AM, Blackwell JR, et al. Muscle metabolic and neuromuscular determinants of fatigue during cycling in different exercise intensity domains. *J Appl Physiol*. 2017;122(3):446-459. doi:10.1152/japplphysiol.00942.2016

20. Burnley M, Doust JH, Vanhatalo A. A 3-min all-out test to determine peak oxygen uptake and the maximal steady state. *Med Sci Sports Exerc*. 2006;38(11):1995-2003. doi:10.1249/01.mss.0000232024.06114.a6

21. Vanhatalo A, Poole DC, DiMenna FJ, Bailey SJ, Jones AM. Muscle fiber recruitment and the slow component of O2 uptake: constant work rate vs. all-out sprint exercise. *Am J Physiol Regul Integr Comp Physiol*. 2011;300(3):R700-7. doi:10.1152/ajpregu.00761.2010

22. Mortensen SP, Dawson E a, Yoshiga CC, et al. Limitations to systemic and locomotor limb muscle oxygen delivery and uptake during maximal exercise in humans. *J Physiol*. 2005;566(Pt 1):273-285. doi:10.1113/jphysiol.2005.086025

23. Mortensen SP, Damsgaard R, Dawson E a, Secher NH, González-Alonso J. Restrictions in systemic and locomotor skeletal muscle perfusion, oxygen supply and VO2 during high-intensity whole-body exercise in humans. *J Physiol*. 2008;586(10):2621-2635. doi:10.1113/jphysiol.2007.149401

24. Hogan MC, Roca J, West JB, Wagner PD. Dissociation of maximal O2 uptake from O2 delivery in canine gastrocnemius in situ. *J Appl Physiol*. 1989;66(3):1219-1226. http://jap.physiology.org/content/jap/66/3/1219.full.pdf. Accessed August 12, 2014.

25. Aaron EA, Seow KC, Johnson BD, Dempsey JA. Oxygen cost of exercise hyperpnea: implications for performance. *J Appl Physiol*. 1992;72(5):1818-1825. http://jap.physiology.org/content/jap/72/5/1818.full.pdf. Accessed August 7, 2014.

26. Poole DC, Ward S a, Gardner GW, Whipp BJ. Metabolic and respiratory profile of the upper limit for prolonged exercise in man. *Ergonomics*. 1988;31(9):1265-

1279. doi:10.1080/00140138808966766

27. Harms CA, Wetter TJ, St Croix CM, Pegelow DF, Dempsey JA. Effects of respiratory muscle work on exercise performance. *J Appl Physiol*. 2000;89(1):131-138. http://jap.physiology.org/content/89/1/131.short. Accessed August 12, 2014.

28. Scharhag-Rosenberger F, Carlsohn A, Lundby C, Schüler S, Mayer F, Scharhag J. Can more than one incremental cycling test be performed within one day? *Eur J Sport Sci*. 2014;14(5):459-467. doi:10.1080/17461391.2013.853208

TABLES

Table 1: Reproducibility of normalized root mean square activity (%RMS_{max}) for four lower limbs muscles during a maximal incremental cycling test.

	ICC	CV (%)	Typical error (percentage points)
VL (%RMS_{max})	0.98	9.8	5.8
VM (%RMS_{max})	0.85	23.5*	15.0*
BF (%RMS_{max})	0.96	5.7	3.2
GM (%RMS_{max})	0.97	6.6	4.8

ICC, Intraclass correlation coefficient; CV, coefficient of variation; VL, *vastus lateralis*; VM *vastus medialis*; BF, *biceps femoris*; GM, *gastrocnemius medialis*. * Removal of one extreme participant would reduce CV to 7.6 and the typical error to 8.1; ICC would increase to 0.98.

Table 2: Physiological data at $\dot{V}O_{2max}$ and P value for Student's *t-tests* between an incremental (INC) and a decremental (DEC) exercise tests.

	INC	DEC	t-test	95% CI	d_z
$\dot{V}O_{2max}$ ($mL.min^{-1}$)	4,139 (657)	4,259 (695)	$t_{(18)} = 2.433$, $p = 0.026$	16 – 224	0.56
HR ($beats.min^{-1}$)	180 (9)	179 (11)	$t_{(18)} = 0.691$, $p = 0.495$	-3 – 1.6	0.16
$\dot{V}_E$ ($L.min^{-1}$)	153.7 (26.7)	160.7 (29.5)	$t_{(18)} = 2.263$, $p = 0.036$	0.5 – 13.5	0.52
BR ($breaths.min^{-1}$)	49.0 (4.8)	54.2 (6.6)	$t_{(10)} = 2.670$, $p = 0.023$	0.8 – 9.5	0.80
V_T (mL)	3.30 (0.53)	3.05 (0.40)	$t_{(10)} = 2.073$, $p = 0.065$	-0.51 – 0.02	0.62
RER	1.13 (0.18)	1.11 (0.14)	$t_{(17)} = 0.945$, $p = 0.357$	-0.05 – 0.02	0.22
$\dot{V}CO_2$ ($mL.min^{-1}$)	4,770 (852)	4,817 (826)	$t_{(18)} = 0.738$, $p = 0.470$	-80 – 177	0.17

$\dot{V}O_{2max}$, maximal oxygen consumption; HR, heart rate; $\dot{V}_E$, ventilation (BTPS); VT, tidal volume; BR, breathing rate; RER, respiratory exchange ratio; $\dot{V}CO_2$, carbon dioxide output.

Table 3: Frequency domain variables of EMG during the INC and DEC tests (at $\dot{V}O_{2max}$).

	INC	DEC	t-statistic	95% CI	d_z
Vastus Lateralis					
Median Frequency (Hz)	44.6 (3.8)	44.2 (4.1)	$t_{(7)} = 1.206$, $p = 0.267$	-1.3 – 0.4	0.43
Mean frequency (Hz)	46.3 (3.6)	45.8 (3.8)	$t_{(7)} = 1.042$, $p = 0.332$	-1.6 – 0.6	0.37
Vastus Medialis					
Median Frequency (Hz)	41.1 (3.1)	41.1 (2.5)	$t_{(8)} = 0.056$, $p = 0.957$	-1.6 – 1.7	0.01
Mean frequency (Hz)	43.2 (3.2)	43.3 (2.4)	$t_{(8)} = 0.104$, $p = 0.919$	-1.3 – 1.5	0.03
Biceps Femoris					
Median Frequency (Hz)	81.9 (16.9)	84.1 (16.5)	$t_{(8)} = 1.935$, $p = 0.101$	-0.6 – 4.8	0.73
Mean frequency (Hz)	100.8 (19.3)	103.3 (19.9)	$t_{(6)} = 1.606$, $p = 0.159$	-1.3 – 6.3	0.60
Gastrocnemius Medialis					
Median Frequency (Hz)	43 (2.5)	42.7 (2.4)	$t_{(6)} = 0.515$, $p = 0.625$	-1.6 – 1.0	0.19
Mean frequency (Hz)	45 (1.8)	45 (2.2)	$t_{(6)} = 0.175$, $p = 0.866$	-0.7 – 0.8	0.06

$\dot{V}O_{2max}$, maximal oxygen consumption.

Table 4: Changes in EMG activity prior to $\dot{V}O_{2max}$ of the incremental test

	@ $\dot{V}O_{2max}$	@ $\dot{V}O_{2max}$ – 20 W	t-statistic	95% CI	d_z
Vastus Lateralis					
RMS (% Sprint)	100.1 (32.4)	92.8 (28.6)	$t_{(6)} = 2.441$, $p = 0.050$	-14.6 – 0.02	0.92
TA (%)	44.3 (17.2)	40 (15.5)	$t_{(6)} = 2.930$, $p = 0.026$	-7.8 – -0.7	1.10
Vastus Medialis					
RMS (% Sprint)	103.3 (40.4)	99.8 (32.1)	$t_{(8)} = 0.906$, $p = 0.391$	-12.4 – 5.4	0.30
TA (%)	49.1 (28.5)	45.5 (27.4)	$t_{(8)} = 3.384$, $p = 0.010$	-6.0 – -1.1	1.06
Biceps Femoris					
RMS (% Sprint)	85.7 (20.5)	76.5 (31.1)	$t_{(6)} = 0.747$, $p = 0.483$	-39.2 – 20.8	0.28
TA (%)	44.0 (5.5)	38.5 (14.2)	$t_{(6)} = 0.993$, $p = 0.359$	-18.9 – 8.0	0.37
Gastrocnemius Medialis					
RMS (% Sprint)	76.5 (24.7)	77.7 (24.4)	$t_{(5)} = 0.232$, $p = 0.825$	-12.4 – 14.8	0.09
TA (%)	31.8 (19.2)	34.1 (19.0)	$t_{(5)} = 0.426$, $p = 0.688$	-11.3 – 15.9	0.17

RMS, root mean square; TA, total activation. All comparisons performed using Student's T-test for paired samples.

FIGURE LEGENDS

Figure 1: Study design diagram. Visits were separated by 2-3 days. See text for detailed description of each testing protocol. Part A tested the $\dot{V}O_{2max}$ between protocols when subjects had never been exposed to a DEC test before. In Part B, one familiarization procedure had been performed on a previous session.

Figure 2: Individual and mean (SD) $\dot{V}O_{2max}$ responses during an incremental and a decremental exercise test. * $p < 0.05$.

Figure 3: Individual and mean (SD) root mean square (RMS) of EMG activity (as a percentage of sprint RMS) and total activation (TA, RMS % multiplied by the fraction of active time within a minute) at $\dot{V}O_{2max}$ during an incremental exercise test and a decremental exercise test. VL, *vastus lateralis (n = 7)*; GM, *gastrocnemius medialis (n = 7);* VM, vastus medialis (n = 9); BF, *biceps femoris (n = 7).*