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## Incremental and decremental cardiopulmonary exercise testing protocols produce similar maximum oxygen uptake in athletes

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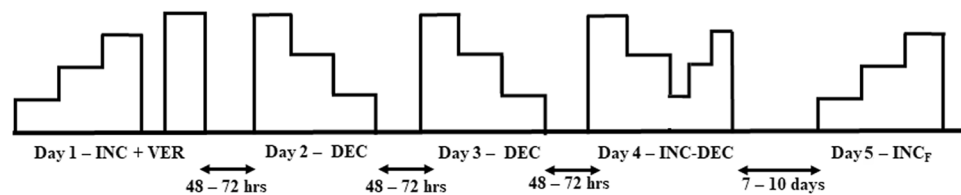
The aim of the study was to evaluate and compare the maximal oxygen uptake ( $\dot{V}O_{2max}$ ) achieved during incremental and decremental protocols in highly trained athletes. Nineteen moderate trained runners and rowers completed, on separate days, (i) an initial incremental  $\dot{V}O_{2max}$  test (INC) on a treadmill, followed by a verification phase (VER); (ii) a familiarization of a decremental test (DEC); (iii) a tailored DEC; (iv) a test with decremental and incremental phases (DEC-INC); (v) and a repeated incremental test (INC<sub>F</sub>). During each test  $\dot{V}O_2$ , carbon dioxide production, ventilation, heart and breath rates and ratings of perceived exertion were measured. No differences were observed in  $\dot{V}O_{2max}$  between INC ( $61.3 \pm 5.2$  ml kg<sup>-1</sup> min<sup>-1</sup>) and DEC ( $61.1 \pm 5.1$  ml kg<sup>-1</sup> min<sup>-1</sup>; average difference of  $-11.58$  ml min<sup>-1</sup>;  $p = 0.831$ ), between INC and DEC-INC ( $60.9 \pm 5.3$  ml kg<sup>-1</sup> min<sup>-1</sup>; average difference of  $-4.8$  ml min<sup>-1</sup>;  $p = 0.942$ ) or between INC and INC<sub>F</sub> ( $60.7 \pm 4.4$  ml kg<sup>-1</sup> min<sup>-1</sup>;  $p = 0.394$ ).  $\dot{V}O_{2max}$  during VER ( $59.8 \pm 5.1$  ml kg<sup>-1</sup> min<sup>-1</sup>) was  $1.50 \pm 2.20$  ml kg<sup>-1</sup> min<sup>-1</sup> lower ( $\sim 2.45\%$ ;  $p = 0.008$ ) compared with values measured during INC. The typical error in the test-to-test changes for evaluating  $\dot{V}O_{2max}$  over the five tests was  $2.4$  ml kg<sup>-1</sup> min<sup>-1</sup> (95% CI  $1.4$ – $3.4$  ml kg<sup>-1</sup> min<sup>-1</sup>). Decremental tests do not elicit higher  $\dot{V}O_{2max}$  than incremental tests in trained runners and rowers, suggesting that a plateau in  $\dot{V}O_2$  during the classic incremental and verification tests represents the maximum ceiling of aerobic power.

The concept of maximal oxygen uptake ( $\dot{V}O_{2max}$ ) was introduced in 1924 after the works of the Nobel laureate Archibald Hill and his colleagues<sup>1</sup>. They observed a linear relationship between workload and oxygen uptake until the  $\dot{V}O_{2max}$  was reached and proposed that the body has a limited capacity to uptake, transport and/or consumption the oxygen, called physiological ceiling for cardiorespiratory fitness<sup>1</sup>. Since then,  $\dot{V}O_{2max}$  has been considered one of the most important indicators of endurance capacity<sup>2</sup> and its determination has become one of the most widely used test procedures in experimental and clinical exercise physiology<sup>3</sup> for testing the cardiorespiratory fitness and performance of athletes, the efficacy of training strategies or ergogenic aids and for quantifying the functional predations of chronic diseases<sup>3,4</sup>.

Because  $\dot{V}O_{2max}$  is an important outcome for both physical performance and health status, test designs that increase the reliability and validity of  $\dot{V}O_{2max}$  determination have widespread applicability. Since the 1970s, the maximal incremental exercise test has become a popular method for establishing  $\dot{V}O_{2max}$ <sup>5</sup>. The  $\dot{V}O_{2max}$  determination typically requires subjects to continue the incremental exercise test until they reach their limit of tolerance<sup>6</sup>. However, Bentley, et al.<sup>7</sup> draw attention to methodological factors that influence physiological parameters such as  $\dot{V}O_{2max}$  in trained endurance athletes during the incremental exercise protocol. In this sense, the application of criteria to assess whether a 'true'  $\dot{V}O_{2max}$  was achieved during an incremental test is still widely discussed by exercise physiologists.

The first and primary criterion traditionally used for establishing that a 'true'  $\dot{V}O_{2max}$  has been reached is when there is no increase in  $\dot{V}O_2$  in response to an increase in work rate at the end of the incremental test: a plateau

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**Figure 1.** Experimental design. First four sessions were separated by 48–72 h between them and the fifth by 7–10 days. *INC* incremental test, *VER* verification phase test, *DEC* decremental test, *DEC-INC* decremental-incremental test, *INC<sub>F</sub>* incremental final.

in  $\dot{V}O_{2max}$ <sup>8–11</sup>. Unfortunately, the  $\dot{V}O_{2max}$  plateau is not always identified in all test individuals<sup>12,13</sup>. Age, modality tested, data processing, physical fitness and test design can influence the incidence of plateaus in  $\dot{V}O_{2max}$ <sup>9,14,15</sup>. In those instances where a  $\dot{V}O$  plateau is not attained as definitive evidence of  $\dot{V}O_{2max}$ , investigators commonly elect to substantiate that  $\dot{V}O_{2max}$  was actually achieved by utilization of secondary criteria assumed to validate  $\dot{V}O_{2max}$ —heart rate (HR)  $\leq 5\%$  of the age-predicted (220-age) maximum, blood lactate concentration  $\geq 8$  mM, or respiratory exchange ratio (RER)  $> 1.00$ , 1.10, or 1.15<sup>16</sup>. Utilization of these criteria can allow for a 30–40% underestimation of the ‘true’  $\dot{V}O_{2max}$  and/or an errant rejection of tests in which subjects had actually achieved their  $\dot{V}O_{2max}$ <sup>16</sup>. To reduce these limitations, a supramaximal verification test, first proposed by Thoden<sup>17</sup> was used in this century to confirm the ‘true’  $\dot{V}O_{2max}$ <sup>16,18–21</sup>. The supramaximal verification test involves a single square-wave bout of exercise performed shortly after the incremental test with a workload higher than the last completed stage of the maximal incremental test. The  $\dot{V}O_{2max}$  results were compared between phases and consistent  $\dot{V}O_{2max}$  values in the incremental and verification phases confirms that a ‘true’  $\dot{V}O_{2max}$  has been attained<sup>10,19–22</sup>.

Beyond the methodological factors, there also remains considerable debate regarding the factors regulating or limiting  $\dot{V}O_{2max}$ <sup>23–25</sup>. The classical model, based on the studies of Hill, et al.<sup>1</sup> and in accordance with the ‘true’  $\dot{V}O_{2max}$  achieved during incremental and verification tests, proposed that  $\dot{V}O_{2max}$  is limited by the maximal cardiac output (cardiac limitation)<sup>26</sup> and the diffusional transport of oxygen out of the muscle microcirculation (muscle limitation)<sup>27</sup>. However, at the beginning of this century, Noakes and Marino<sup>28</sup> introduced another theory into this discussion, which states that the cardiovascular system never reaches a limit of work and that  $\dot{V}O_{2max}$  is regulated, rather than limited, by the number of motor units recruited in the exercising limbs, which is always submaximal. This model proposes that the central nervous system (a central governor) controls the circulation during severe exercise and that there is always both cardiovascular and neuromuscular reserve upon exhaustion during incremental exercise<sup>23</sup>; however, the brain stops the exercise to prevent catastrophic failure in the body system<sup>28</sup>.

Thus, the ‘central governor’ model, supported by the experimental design with decremental tests of Beltrami, et al.<sup>29</sup> suggests that the  $\dot{V}O_{2max}$  achieved during incremental exercise is not the ‘true’  $\dot{V}O_{2max}$ . Beltrami, et al.<sup>29</sup> supported the decremental test design with: (i) an incremental test may cause more anticipatory stress, which may lead to a difference in blood flow response; (ii) a decremental test, with workload progressively easier, might relax brain controls directing the termination of exercise<sup>28,30</sup>; (iii) evidence suggests that submaximal decremental protocols produce higher-than-expected  $\dot{V}O_2$  values compared with a similar power output during an incremental protocol<sup>31,32</sup>. In fact, Beltrami, et al.<sup>29</sup> showed that decremental protocols elicit significantly higher  $\dot{V}O_{2max}$  values (~4%) than incremental protocol in subjects involved in running and cross-country skiing training. On the other hand, Taylor, et al.<sup>33</sup>, using similar decremental tests in running and triathlon training subjects, did not report differences in the  $\dot{V}O_{2max}$  achieved during incremental or decremental tests, and both protocols elicited a similar cardiovascular response. These results showed that  $\dot{V}O_{2max}$  determination is still challenging, not only from a methodological point of view but also from the factors limiting  $\dot{V}O_{2max}$ , although it is widely used and well established. Considering the scarce literature on decremental protocols for testing the regulatory factors of  $\dot{V}O_{2max}$ , the aim of this study was to evaluate and compare the  $\dot{V}O_{2max}$  achieved during incremental and decremental protocols in trained athletes.

## Methods

**Participants.** The study included 19 moderate trained men who competed in the junior category at a national or, in some cases, international level (15 distance runners and 4 rowers; age  $17.4 \pm 1.0$  years, body mass  $69.2 \pm 6.3$  kg, height  $174.6 \pm 4.6$  cm and body fat  $8.3 \pm 1.6\%$ ) with at least 6 months of uninterrupted training. The rowers were former runners and they had running as part of their training routine. The participants were injury-free and did not use any controlled drugs or nutritional supplementation during the experimental protocols. All procedures were approved by the university’s Ethics Committee (51127515.8.0000.5284) and were conducted in accordance with the Declaration of Helsinki. The subjects were kept informed of experimental procedures and risks and signed informed consent before participation in the study. The study followed the STROBE (STrengthening the Reporting of OBServational studies in Epidemiology). All trial sessions took place in one laboratory using the same equipment.

**Experimental design.** The participants visited the university laboratory on five occasions (Fig. 1) to complete the study. The sessions were separated by 48–72 h between the first four visits and by 7–10 days between

the fourth and the fifth visit. All tests were performed using a motored treadmill (Super ATL, Inbrasport, Porto Alegre, RS, Brazil), a gas analyser (Metalyzer II, Cortex, Leipzig, Germany) that was calibrated before each test according to the manufacturer's instructions, a heart rate monitor (Polar S810i, Kempe, Finland) and a Borg perception effort scale<sup>34</sup>.

On the first visit, the participants performed an incremental test (INC) until volitional exhaustion, and after 15 min a verification phase test (VER) was performed. A familiarization decremental test (DEC) was performed on the second visit and a tailored DEC on the third visit. On the fourth visit, a test with decremental and incremental phases (DEC-INC) was applied. Finally, a repeated incremental test was applied on the fifth visit (INC<sub>F</sub>).

The participants were instructed to avoid hard training sessions during the data collection period (15–20 days) and not to ingest caffeine or any kind of stimulant for 6 h before the tests. Tests were scheduled at the same time of the day (afternoon; between 3 and 5 p.m.) and laboratory conditions were stable for the duration of the study (ambient temperature 20–22 °C, relative air humidity 50–60%). All tests were performed on a motor-driven treadmill with a constant inclination of 5% (to ensure that participants do not exceed the capacity of the treadmill). All trial sessions were completed in 15–20 days by each participant. All tests were preceded by a 5-min warm-up at 10 km h<sup>-1</sup> with no treadmill inclination and participants were verbally encouraged throughout each test.

**Incremental test (INC) and verification phase test (VER).** INC started at 9 km h<sup>-1</sup> and 5% grade and the speed was increased by 1 km h<sup>-1</sup> every minute until volitional exhaustion. Fifteen minutes after the end of INC in the first session the participants performed VER to confirm the values of  $\dot{V}O_{2max}$ . VER began at 10 km h<sup>-1</sup> and 5% grade for 1 min and then the speed was increased to 1 km h<sup>-1</sup> above the maximum speed reached in the previous INC. Participants were instructed to run at that speed for as long as they could. During the interval between INC and VER they were advised to rest or walk. The value of  $\dot{V}O_{2max}$  was considered the highest after a mean of 20 s (with the data interpolated on a second by second basis and then averaged to retrieve the  $\dot{V}O_{2max}$  value). INC was repeated on the fifth visit (INC<sub>F</sub>) without VER in order to mislead the training effect on  $\dot{V}O_{2max}$  during the experimental protocol.

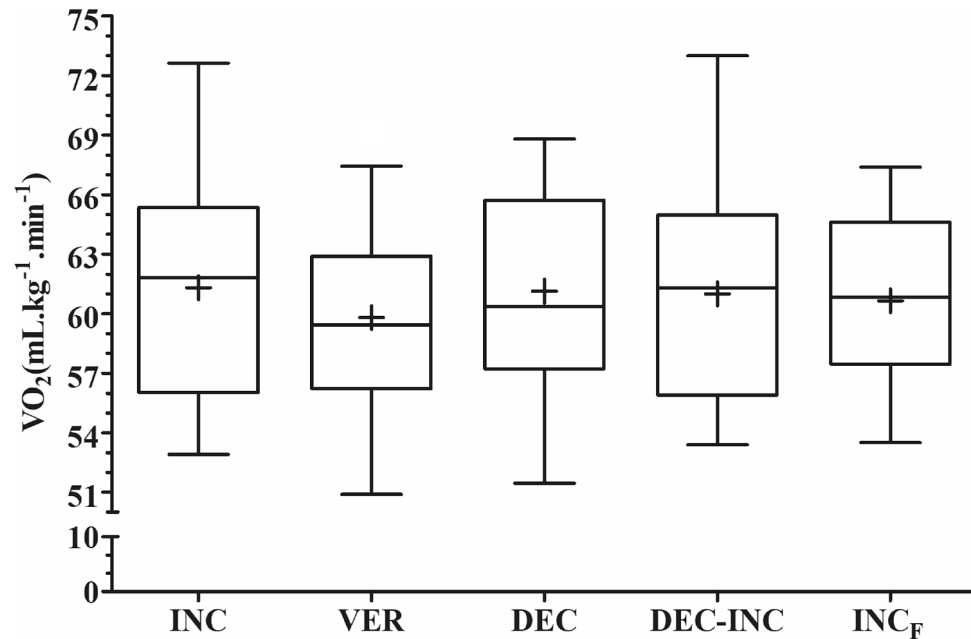
**Decremental test (DEC).** DEC protocols (familiarization and tailored) were established as a function of the result from the first session (INC + VER). The familiarization DEC was performed at the second visit. The first stage consisted of a graded increase in the speed of the treadmill until it reached that in VER (2 min at 9–10 km h<sup>-1</sup>, 1.5 min at 12–13 km h<sup>-1</sup> and 30 s at 15.5–16.5 km h<sup>-1</sup>). This graded increase in speed was included to diminish the gap in speed between the warm-up and the high-intensity start of the test. After reaching the VER speed, participants ran for 60% of the time that each had managed during VER (usually around 1 min). After this stage, speed was decreased by 1 km h<sup>-1</sup> and maintained for 30 s. The speed in the following stages had consecutive decreases of 0.5 km h<sup>-1</sup> that were maintained for 30, 45, 60, 90 and 120 s, respectively. The tailored DEC was performed at the third visit, the duration of the stages being determined by the individual reaction of the participant to the familiarization DEC. Some adjustments (increase or decrease in stage duration) were made to ensure that DEC was longer than 5 min. The value of  $\dot{V}O_{2max}$  was considered the highest after a mean of 20 s. No physiological data were measured during the familiarization DEC.

**Decremental and incremental test (DEC-INC).** DEC-INC was established as a function of the result from the DEC session. The first phase was similar to DEC and lasted until reaching the speed for  $\dot{V}O_{2max}$  achieved during DEC. After reaching this speed, the second phase of DEC-INC consisted of consecutive 10 s stages with increments of 0.5 km h<sup>-1</sup> until volitional exhaustion. The value of  $\dot{V}O_{2max}$  was considered the highest after a mean of 20 s.

**Instruments and data handling.** All physiological data were collected within a fixed time of 10 s, exported from the analyser software into Excel spreadsheets. During INC, data were collected until the end of the final completed stage. Data collected during VER, DEC and DEC-INC were considered up to the final time (at least 30 s) the respective test had been collected. This was done to ensure that comparison between INC and VER was done at two different workloads, as required to define a plateau in  $\dot{V}O_2$ . The maximum test confirmation criteria for INC and INC<sub>F</sub> were: (a) the volitional exhaustion and a plateau in  $\dot{V}O_2$ ; (b) when the plateau in  $\dot{V}O_2$  was not achieved, the volitional exhaustion and one of the following criteria: heart rate (HR)  $\leq 5\%$  of the age-predicted (220-age) maximum or respiratory exchange ratio (RER)  $> 1.00$ . A plateau in  $\dot{V}O_2$  during INC was accepted if the change in  $\dot{V}O_2$  during the highest 30-s interval between the two final stages of the test was less than half of the normal stage-to-stage change in  $\dot{V}O_2$  during the initial (linear) parts of the tests for each subject<sup>29</sup>. The average stage-to-stage difference in  $\dot{V}O_2$  for all participants was calculated as  $320 \pm 66$  ml min<sup>-1</sup>, so the plateau phenomenon was defined as a change in  $\dot{V}O_2 < 160 \pm 33$  ml min<sup>-1</sup> (or an average of  $\pm 2.2$  ml kg<sup>-1</sup> min<sup>-1</sup>, considering the average body mass of the participants) between the two final stages of the test. The same criterion was used to define a plateau in  $\dot{V}O_2$  between the  $\dot{V}O_{2max}$  values measured during INC and VER, because VER was performed at one stage higher than the maximal stage completed during INC<sup>29</sup>.

Considering the stage when  $\dot{V}O_{2max}$  was reached for each test, minute ventilation (VE), breathing rate (BR), respiratory exchange ratio (RER), heart rate (HR) and rating of perceived exertion (RPE) were evaluated.

**Statistical analysis.** Results are expressed as the mean  $\pm$  standard deviation (SD). The normality of the data was tested and confirmed by Shapiro–Wilk's test ( $p > 0.05$ ), which allowed the use of parametric statistics. A one-way repeated measures ANOVA was used to compare the speed and respiratory and psychological variables at  $\dot{V}O_{2max}$  between the proposed tests (INC, VER, DEC, DEC-INC and INC<sub>F</sub>). Compound sphericity was verified by the Mauchly test. When the assumption of sphericity was not met, the significance of F-ratios was



**Figure 2.** Maximal oxygen consumption ( $\dot{V}O_{2max}$ ) during the five different protocols. *INC* incremental test, *VER* verification phase test, *DEC* decremental test, *DEC-INC* decremental–incremental test, *INC<sub>F</sub>* final incremental test. No statistically significant differences were observed between protocols.

adjusted according to the Greenhouse–Geisser procedure. Tukey's post-hoc test with the Bonferroni adjustment was applied in the event of significance. Sample size was determined using G\*Power version 3.1.3<sup>35</sup>, based on the difference in  $\dot{V}O_{2max}$  between *INC* and *VER* during a pilot study with  $n=5$  and the  $\dot{V}O_{2max}$  presented by Beltrami, et al.<sup>29</sup>. Considering the effect size (ES) achieved ( $\sim 0.75$ ), alpha error of 0.05 and power ( $1 - \beta$ ) of 0.80, the required sample size was  $n=16$ . All analyses were conducted using the Statistical Package for Social Science software, version 21.0 (SPSS Inc., Chicago, IL, USA), and the significance level was set at  $p < 0.05$ . The typical error in the test-to-test changes in  $\dot{V}O_{2max}$  was calculated using an Excel spreadsheet that calculates reliability statistics for consecutive pairs of trial sessions<sup>36</sup>.

**Ethics approval.** National Research Ethics Committee Brazil (CAEE 51127515.8.0000.5284).

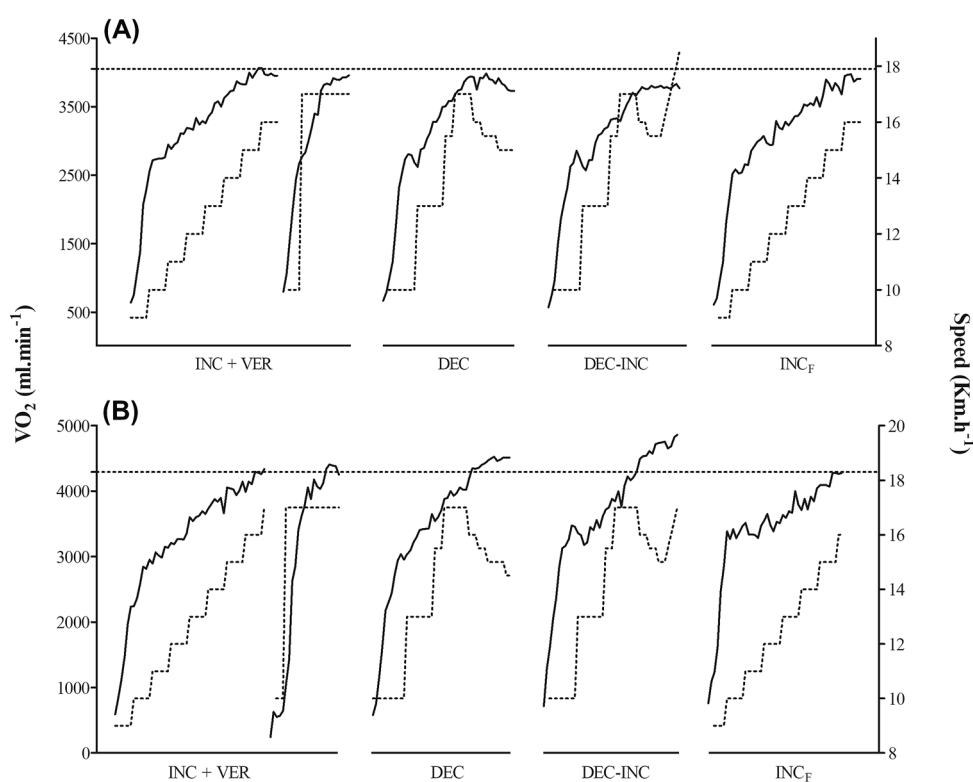
## Results

The plateau phenomenon in  $\dot{V}O_2$  was observed in 13 participants during *INC*, 7 participants during *VER* and 12 participants during *INC<sub>F</sub>*. When the *INC* and *VER* protocols were combined, 17 participants achieved the plateau phenomenon. Figure 2 shows the  $\dot{V}O_{2max}$  achieved during all five protocols. The protocol intervention did not elicit statistically significant changes in  $\dot{V}O_{2max}$  values ( $F(60, 4) = 0.80$ ,  $p = 0.528$ ,  $ES = 0.05$ ). Post hoc analysis with a Bonferroni adjustment revealed that there were no differences in  $\dot{V}O_{2max}$  between *INC* and *INC<sub>F</sub>* ( $61.3 \pm 5.2$  vs.  $60.7 \pm 4.4$   $\text{ml kg}^{-1} \text{min}^{-1}$ , respectively;  $p = 1.000$ ), indicating no familiarization between tests. No differences in  $\dot{V}O_{2max}$  were observed with post hoc analysis between *INC* and *VER* ( $59.8 \pm 5.1$   $\text{ml kg}^{-1} \text{min}^{-1}$ ; average difference of  $1.50 \pm 2.20$   $\text{ml kg}^{-1} \text{min}^{-1}$ ,  $\sim 2.45\%$ ;  $p = 0.127$ ), *INC* and *DEC* ( $61.1 \pm 5.1$   $\text{ml kg}^{-1} \text{min}^{-1}$ ; average difference of  $0.17 \pm 3.30$   $\text{ml kg}^{-1} \text{min}^{-1}$ ,  $\sim 11.58$   $\text{ml min}^{-1}$ ;  $p = 0.831$ ) or between *INC* and *DEC-INC* ( $60.9 \pm 5.3$   $\text{ml kg}^{-1} \text{min}^{-1}$ ; average difference of  $\sim 0.07 \pm 4.20$   $\text{ml kg}^{-1} \text{min}^{-1}$ ,  $4.8$   $\text{ml min}^{-1}$ ;  $p = 0.942$ ), which is 73% lower than the threshold for a  $\dot{V}O_2$  plateau during *INC* ( $2.2$   $\text{ml kg}^{-1} \text{min}^{-1}$ ,  $159.9$   $\text{ml min}^{-1}$ ). Considering these protocols from the same objective, to evaluate  $\dot{V}O_{2max}$ , the typical error in test-to-test changes for  $\dot{V}O_{2max}$  over the five tests was  $2.4$   $\text{ml kg}^{-1} \text{min}^{-1}$  (95% CI =  $1.4$ – $3.4$   $\text{ml kg}^{-1} \text{min}^{-1}$ ).

All physiological variables evaluated during the protocols are listed in Table 1. The protocol intervention did not elicit statistically significant changes in VE [ $F(60, 4) = 2.05$ ,  $p = 0.099$ ,  $ES = 0.12$ ] and RPE [ $F(36, 3) = 2.92$ ,  $p = 0.057$ ,  $ES = 0.20$ ] values. Statistically significant changes were observed in BR [ $F(60, 4) = 4.00$ ,  $p = 0.006$ ,  $ES = 0.21$ ] and HR [ $F(36, 3) = 10.05$ ,  $p \leq 0.0005$ ,  $ES = 0.46$ ], with lower BR values in *INC<sub>F</sub>* than in *INC* and lower HR values in *DEC* and *DEC-INC* than in *INC* and *INC<sub>F</sub>*. The protocol intervention elicited statistically significant changes in RER values [ $F(60, 4) = 19.44$ ,  $p \leq 0.0005$ ,  $ES = 0.56$ ], with the lower RER achieved during *VER* and the highest RER during *DEC* and *DEC-INC*. The *INC* and *INC<sub>F</sub>* did not present statistically significant differences in RER. The protocol also elicited statistically significant changes in  $v\dot{V}O_{2max}$  [ $F(60, 4) = 9.20$ ,  $p \leq 0.0005$ ,  $ES = 0.38$ ] and TTE [ $F(60, 4) = 53.78$ ,  $p \leq 0.0005$ ,  $ES = 0.78$ ], with the *VER* protocol presented higher  $v\dot{V}O_{2max}$  and lower TTE than all other protocols. The *DEC* protocol also presented lower TTE than *INC* and *INC<sub>F</sub>*.

	INC	VER	DEC	DEC-INC	INC <sub>F</sub>
$\dot{V}O_{2max}$ (L min <sup>-1</sup> )	4.23 ± 0.58	4.13 ± 0.57	4.22 ± 0.55	4.23 ± 0.55	4.15 ± 0.43
VE (L min <sup>-1</sup> )	119.9 ± 14.7	118.7 ± 14.9	116.6 ± 12.9	116.7 ± 14.4	111.8 ± 12.1
BR (BR min <sup>-1</sup> )	64.6 ± 6.1	65.9 ± 7.3	61.0 ± 7.1	61.9 ± 7.4	59.6 ± 6.9 <sup>†</sup>
RER	1.09 ± 0.05	1.00 ± 0.07 <sup>*</sup>	1.14 ± 0.03 <sup>*†</sup>	1.16 ± 0.87 <sup>*†</sup>	1.10 ± 0.05 <sup>†‡</sup>
HR (b min <sup>-1</sup> )	197.3 ± 7.3	–	193.1 ± 5.7 <sup>*</sup>	190.8 ± 8.0 <sup>*</sup>	195.7 ± 8.0 <sup>§</sup>
RPE	16.8 ± 2.2	–	14.8 ± 3.8	15.5 ± 4.0	17.2 ± 2.6
TTE (s)	477 ± 72	130 ± 24 <sup>*</sup>	393 ± 43 <sup>*†</sup>	403 ± 60 <sup>†</sup>	477 ± 69 <sup>†‡</sup>
$v\dot{V}O_{2max}$ (km h <sup>-1</sup> )	15.8 ± 1.1	16.8 ± 1.1 <sup>*</sup>	15.7 ± 1.7 <sup>†</sup>	16.0 ± 1.2 <sup>†</sup>	16.0 ± 1.1 <sup>†</sup>

**Table 1.** Physiological variables achieved at  $\dot{V}O_{2max}$  during the different protocols.  $\dot{V}O_{2max}$  maximal oxygen consumption, VE minute ventilation, BR breath rate, RER respiratory exchange rate, HR heart rate, RPE rating of perceived exertion, TTE time to exhaustion,  $v\dot{V}O_{2max}$  intensity at  $\dot{V}O_{2max}$ , INC incremental test, VER verification phase test, DEC decremental test, DEC-INC incremental test, INC<sub>F</sub> final incremental test. \* $p < 0.05$  for INC; <sup>†</sup> $p < 0.05$  for VER; <sup>\*</sup> $p < 0.05$  for DEC; <sup>†</sup> $p < 0.05$  for DEC-INC; – did not recorded.



**Figure 3.** Maximal oxygen consumption ( $\dot{V}O_{2max}$ ) and speed treadmill during the five different protocols for a participant who presented similar values of  $\dot{V}O_{2max}$  between INC and DEC (A) and a participant who presented very different values of  $\dot{V}O_{2max}$  between INC and DEC (B).

A graphical illustration of the relationship between  $\dot{V}O_2$  and speed in the different trial sessions for two volunteers is presented in Fig. 3: (A) a participant who presented very similar values of  $\dot{V}O_{2max}$  between protocols; and (B) a participant who presented different values of  $\dot{V}O_{2max}$  between INC and DEC or DEC-INC. When an individual analysis of the sampled subjects was performed, only six achieved a  $\dot{V}O_{2max}$  during DEC and DEC-INC (three are the same subjects) that was  $160 \pm 33$  ml min<sup>-1</sup> (plateau phenomenon calculated in INC) greater than the  $\dot{V}O_{2max}$  achieved during INC. None of the subjects presented a  $\dot{V}O_{2max}$  during VER that was  $160 \pm 33$  ml min<sup>-1</sup> greater than the  $\dot{V}O_{2max}$  achieved during INC. Of the sample that did not develop the plateau phenomenon (six subjects), only two achieved a  $\dot{V}O_{2max}$  during DEC and DEC-INC (the same subjects) that was  $160 \pm 33$  ml min<sup>-1</sup> greater than the  $\dot{V}O_{2max}$  achieved during INC. However, no difference in  $\dot{V}O_{2max}$  was observed between INC and DEC when only the six subjects were evaluated ( $n = 6$ ; INC =  $61.4 \pm 4.8$  ml kg<sup>-1</sup> min<sup>-1</sup>; DEC =  $62.9 \pm 5.0$  ml kg<sup>-1</sup> min<sup>-1</sup>;  $p = 0.160$ ). Individual  $\dot{V}O_{2max}$  response results can be consulted in the Supplementary Information.



## Discussion

The results of the present study showed that, regardless of the test performed, the  $\dot{V}O_{2max}$  achieved in each test was very similar and its difference was smaller than the value of the plateau phenomenon between the last two stages of INC or even the measurement error in test-to-test changes. That is, contrary to the study by Beltrami, et al.<sup>29</sup> no significant differences were found regarding the  $\dot{V}O_{2max}$  values between INC and DEC. Moreover, the test proposed by us with decremental and incremental stages (DEC-INC) also showed no difference from INC. Thus, although more recently the theory of the plateau phenomenon has been criticized<sup>37</sup>, the results of the present study show that the plateau obtained at the time of exhaustion during a traditional test (i.e. INC) may infer the maximum ceiling of cardiorespiratory capacity and thus remain in exercise, as proposed by the classical theory<sup>1,38,39</sup>.

In an attempt to elucidate the validity of a cardiopulmonary test to assess maximal cardiorespiratory capacity, in addition to the plateau phenomenon, a series of validation criteria have been proposed in the literature, such as expected values of HR, RER and blood lactate concentrations<sup>3</sup>. In addition to those already cited, in order to improve the reliability of determining  $\dot{V}O_{2max}$ , VER has been proposed<sup>18</sup>. This test requires the participant to perform the exercise for as long as possible at a constant intensity above that achieved in INC. Therefore, even with increased intensity, if the  $\dot{V}O_{2max}$  value is not higher than that found during INC it is assumed that the maximum aerobic power ceiling was reached during both tests<sup>10,40,41</sup>. From this application, the frequency with which the plateau was identified increased, strengthening the theory that discusses this phenomenon<sup>41</sup> (when the INC and VER protocols were combined, 17 of the 19 participants achieved the plateau phenomenon).

Evidence using VER as an INC validity criterion has controversial results. At some point VER will identify different responses to INC depending on a series of factors (participants, tests performed, equipment used, etc.). Murias, et al.<sup>42</sup> used VER (105%) to analyse the effectiveness of INC on a ramp format in young and old individuals. No significant differences were found between the two tests in the two populations, thus, although VER may be very seductive in finding different values, in that study it did not present any additional validation. Furthermore, Bhammar, et al.<sup>43</sup> when comparing obese and non-obese children, found that the  $\dot{V}O_{2max}$  values in VER (105% of  $\dot{V}O_{2max}$  during INC) were higher by approximately 6% and 10%, respectively. Barker, et al.<sup>44</sup> also found that supramaximal testing at 105% of the power output achieved during ramp exercise did not increase the  $\dot{V}O_{2max}$  achieved compared to the ramp test, thus suggesting the achievement of a true  $\dot{V}O_{2max}$  during the initial ramp test for young people. However, the authors also concluded that the adherence to commonly used secondary criteria to validate a maximal effort (expected values of HR, RER and blood lactate concentration) in young people can result in either a submaximal  $\dot{V}O_{2max}$  or a rejection of a participant's  $\dot{V}O_{2max}$  score despite a plateau being evident. Finally, Beltrami, et al.<sup>29</sup> applied VER (110% of  $\dot{V}O_{2max}$  during INC) to highly trained individuals ( $\dot{V}O_{2max} = 61.3 \text{ ml kg}^{-1} \text{ min}^{-1}$ ) and also found no significant differences, with very small variations between INC and VER. In the present study, the  $\dot{V}O_{2max}$  during VER (110% of  $\dot{V}O_{2max}$  during INC) were not significantly different than during INC ( $\dot{V}O_{2max}$  values during VER were 3% lower;  $p = 0.127$ ; clinically irrelevant). It is clear that during INC the participants had reached the maximum ceiling of cardiorespiratory fitness, which was confirmed by the VER protocol. It is important to highlight that of the 6 participants that did not reach the plateau in  $\dot{V}O_{2max}$  values during the INC, 3 reached during the VER protocol. These data sustained, once again, that the  $\dot{V}O_{2max}$  achieved during INC is the maximum ceiling of cardiorespiratory fitness.

DEC was used by Beltrami, et al.<sup>29</sup> to break with the traditional tests for determination of aerobic power and to evaluate the validity of the traditional INC in determining the maximum ceiling of cardiorespiratory fitness. Beltrami, et al.<sup>29</sup> demonstrated that in DEC, their participants achieved significantly higher  $\dot{V}O_{2max}$  values (4.4%) than those found when applying the traditional INC. Although the study was cited as being innovative as a function of protocol and approach, previous studies, even without the use of DEC as a tool for determining  $\dot{V}O_{2max}$ , are essential for understanding the phenomenon behind the oxygen uptake response to this stimulus<sup>31,33,45-47</sup>. The results of the present study show no differences between INC and DEC in the  $\dot{V}O_{2max}$  achieved, or in the test developed by our research group, DEC-INC, which sought an increase in  $\dot{V}O_{2max}$  with increasing oxygen uptake intensity after its decrease. We did not discard methodological differences between our study and the research of Beltrami, et al.<sup>29</sup>, mainly the difference in the gas analyzer and the difference between the procedures for collection and analysis of respiratory data. These results are in agreement with those obtained by Taylor, et al.<sup>33</sup>, who briefly compared the values of  $\dot{V}O_{2max}$ , cardiac output and systolic volume obtained by means of INC and DEC in very well-trained triathletes and runners, a population similar to that of the present study and also of Beltrami, et al.<sup>29</sup>. Taylor, et al.<sup>33</sup> also found no significant differences between INC and DEC in maximal oxygen uptake values ( $57.29 \pm 8.94 \text{ ml kg}^{-1} \text{ min}^{-1}$  vs.  $60.82 \pm 8.49 \text{ ml kg}^{-1} \text{ min}^{-1}$ , respectively) or in cardiocirculatory values. They also concluded that DEC was not capable of causing higher values of maximal oxygen uptake but may be an alternative for this population. We do not share the same view, as DEC exhibits the same possible obstacles as INC (i.e. the need to establish initial loads, the duration of stages and the decrease in intensity) and is an 'open-ended' test<sup>48</sup>. In addition, due to the need to achieve high intensities in order to decrease intensity later, the error in high intensity selection is much higher than the error in intensity selection for INC. Thus, we have not assumed DEC to be a good alternative for determining  $\dot{V}O_{2max}$ , although it may be explored in the future.

Even with the emergence and acceptance of VER as a tool to try to validate the maximum exercise condition maintained by an individual, the debate about other factors that permeate the phenomenon of  $\dot{V}O_{2max}$  remains intense. Among the most common theory is that the cardiovascular system limits  $\dot{V}O_{2max}$  in a maximal or supramaximal situation<sup>23,49</sup>. According to Elliott, et al.<sup>23</sup> the evidence is controversial. For instance, some studies show that cardiac output stabilizes at maximal and supramaximal loads, whereas others found that cardiac output does not stabilize in athletes. Barker et al.<sup>44</sup> demonstrated in young people that the changes in the components of the Fick equation, that is maximal cardiac output and  $O_2$  extraction, were similar during ramp and supramaximal exercise, further supporting the notion that a true  $\dot{V}O_{2max}$  was recorded during the ramp test. However, there

is still little evidence related to the cardiovascular system in DEC. Taylor, et al.<sup>33</sup> demonstrated no differences in maximal cardiac output and maximal systolic volume between INC and DEC, despite a tendency for higher values in INC and lower HR values in DEC. Interestingly, in the present study, the  $\dot{V}O_{2\max}$  achieved in DEC was obtained at intensities similar to those in INC (already in the descending phase of the test) but with lower TTE and lower values of HR and RER, demonstrating that there could still be a cardiorespiratory reserve but it was not translated into higher values of  $\dot{V}O_{2\max}$ . Another front explored is the convection property of  $O_2$  in tissues, as well as its extraction of blood from the muscles<sup>50</sup>.

We cannot fail to mention that the results for identifying or not the ceiling effect found by different studies can be partially explained by methodological differences (e.g.; chronological age, sex, training level, training status, type of exercise, sports modality, ergometer, criteria for interruption and confirmation of the maximum test, criterion for plateau, motivation strategy, exercise protocol,  $\dot{V}O_2$  sampling frequency, statistical analysis). Studies that used breath by breath analysis have a greater dispersion of  $\dot{V}O_2$  data, which makes it difficult to identify a plateau. On the contrary, data collected every 10 s and analyzed in an average of 30 s (as in our study) decrease the variability of  $\dot{V}O_2$  data, facilitating the identification of a plateau. On the other hand, there are few studies that make clear the criteria for interrupting the maximum test, as well as the criteria used to confirm the maximum test. This can really generate discrepant results and must be taken into consideration when comparisons are made.

According to Hill's classical theory (1924), running and rowing athletes reached the maximum ceiling of aerobic power by conventional testing (similar  $\dot{V}O_{2\max}$  in INC, VER, DEC, DEC-INC and INC<sub>F</sub>), in contrast to other studies<sup>23,29</sup>, that the  $\dot{V}O_{2\max}$  reached during conventional tests was lower than unconventional decremental tests. It is important to highlight that these results were achieved by trained athletes, with the possibility of different results in untrained individuals, since the participation of the different mechanisms behind the onset of fatigue/exhaustion is different depending on the training state. According to this study, the verification phase after the incremental protocol for athletes it's not necessary; however, since not all athletes reach the  $\dot{V}O_{2\max}$  plateau during the incremental protocol, we recommend the verification phase when the excellence is sought (the chances that all athletes reach the plateau in  $\dot{V}O_{2\max}$  increases considerably when INC and VER are combined. Future research should aim to design a DEC protocol for non-athletic populations.

In summary, decremental tests do not elicit higher  $\dot{V}O_{2\max}$  than incremental tests in trained runners and rowers, suggesting that a plateau in  $\dot{V}O_2$  during the classic incremental and verification tests represents the maximum ceiling of aerobic power.

## Data availability

The views expressed in this publication are those of the author(s) and not necessarily those of the funders. All authors had full access to all of the data (including statistical reports and tables) in the study and can take responsibility for the integrity of the data and the accuracy of the data analysis. *Data sharing statement* Data are not publicly available, but applications for data sharing can be made. For enquiries, please contact NMFS (nunofrade@gmail.com).

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## Author contributions

All authors conceived and designed the research. N.M.F.S., G.M.S., P.L.R.A.P. and D.M.R. contributed to the data collection and analysis. N.M.F.S. and D.R.B. wrote the manuscript and prepared the figures and tables. G.M.S., P.L.R.A.P. and D.M.R. contributed edits, feedback, and suggestions for the manuscript. All authors reviewed and approved the manuscript.



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## Competing interests

The authors declare no competing interests.

## Additional information

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