Influence of endurance exercise on respiratory muscle performance

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ABSTRACT


Purpose: During high-intensity, exhaustive, constant-load exercise above 85% of maximal oxygen consumption, the diaphragm of healthy subjects can fatigue. Although a decrease in trans-diaphragmatic pressure is the most objective measure of diaphragmatic fatigue, possible extra-diaphragmatic muscle fatigue would not be detected by this method. The aim of the present study was to investigate the impact of exhaustive, constant-load cycling exercise at different intensities on global respiratory performance determined by the time to exhaustion while breathing against a constant resistance. Methods: Ten healthy, male subjects performed an exhaustive cycling endurance test at 65, 75, 85, and 95% of peak oxygen consumption (\(\dot{V}_{\text{O}_2}\)peak). Before cycling (t0) as well as at 10 min (t10) and 45 min (t45) after cycling, respiratory performance was determined. Results: Breathing endurance was equivalently reduced after exhaustive cycling at either 65% (8.4±4.1 min [t0] vs 3.9±2.8 min [t10]), 75% (9.9±6.1 vs 4.4±2.8 min), 85% (9.3±6.0 vs 3.8±2.9 min), or 95% \(\dot{V}_{\text{O}_2}\)peak (8.5±5.1 vs 4.0±2.5 min) and, therefore, was independent of exercise intensity. Conclusion: This result contradicts previous findings, possibly due to the fact that extra-diaphragmatic muscles are tested in addition to the diaphragm during resistive breathing. Key Words: RESISTIVE BREATHING, RESPIRATORY MUSCLE FATIGUE

During high-intensity, exhaustive, constant-load running or cycling exercise above 85% \(\dot{V}_{\text{O}_2}\)max (1-3,12) or 80% \(W_{\text{max}}\) (18), the diaphragm of healthy subjects can fatigue—as shown by a reduction of trans-diaphragmatic twitch pressure (Pdi,tw) during electrical or magnetic stimulation of the phrenic nerves. At intensities of 70–75% \(W_{\text{max}}\), Pdi,tw was reduced in 9 of 14 subjects only (17). In general, the higher the intensity of exercise, the larger the diaphragmatic fatigue (12)—even in the face of shorter exercise durations at higher intensities.

Although the measurement of Pdi,tw is certainly the most objective measure of diaphragmatic fatigue, there are some limitations to this technique. On the one hand, it is an exclusive measure of diaphragmatic fatigue neglecting possible extra-diaphragmatic inspiratory muscle fatigue and on the other hand, this technique is laboratory-bound and somewhat “invasive.”

In contrast, breathing against a constant resistance until exhaustion, an easy to use and “noninvasive” technique involving most of the inspiratory muscles, has been used in the past to measure global inspiratory muscle fatigue: Ker and Schultz (13) had their subjects breathe to exhaustion against a constant resistive load before and after completion of an ultramarathon. The maximal breathing endurance time remained reduced even 3 d after the ultramarathon. In a recent study, we showed that respiratory performance was reduced by 43% after exhaustive cycling at 85% \(\dot{V}_{\text{O}_2}\)peak (21), a workload at which diaphragmatic fatigue had previously been demonstrated (12). The 43% reduction in breathing endurance time was larger than the reduction in Pdi,tw (8–32%) reported by Johnson et al. (12) after exercise at similar workloads. This difference might result from extra-diaphragmatic fatigue which is measured during resistive breathing.

The aim of the present study was to investigate the impact of exhaustive, constant-load cycling exercise at 65, 75, 85, and 95% \(\dot{V}_{\text{O}_2}\)peak on respiratory muscle performance as determined by the maximal breathing endurance time in a constant-load resistive breathing test. Additionally, we measured blood lactate concentration, pH, serum potassium concentration, and core body temperature, factors known to affect skeletal muscle contractility (6,9,11) and performance (14). We hypothesized that the decrease in respiratory performance after exercise would correlate with exercise intensity as does the decrease in Pdi,tw (12) but that this correlation would possibly have a different slope due to the measurement of both extra-diaphragmatic muscle performance and diaphragmatic muscle performance.

METHODS

Subjects. Ten healthy, nonsmoking, male subjects (study group) participated in the main study. Their average...
age was 29 ± 4 yr, their height was 181 ± 5 cm, and their weight was 72 ± 7 kg. They were physically fit (VO₂peak 60 ± 4 mL·kg⁻¹·min⁻¹) and had normal lung function (Table 1). An additional group of 10 subjects (control group) performed two series of control experiments. Their average age was 26 ± 4 yr, their height was 183 ± 6 cm, and their weight was 71 ± 7 kg. They were physically fit and had normal lung function (Table 1). The two groups had similar physical and lung function characteristics. Informed written consent was obtained from each subject and the study protocol was approved by the Ethics Committee of Physiology and Pharmacology Departments at the University of Zurich.

Subjects were asked to abstain from caffeine intake for at least 2 h before each test, as caffeine increases breathing endurance during loaded breathing tests (22) and attenuates the exercise-induced increase in plasma potassium levels (16). Subjects were also instructed to keep their personal training schedule constant throughout the study protocol and to perform no strenuous workouts the day before a test.

**Equipment.** Vital capacity (VC), forced expiratory volume in 1 s (FEV₁), peak expiratory flow (PEF), maximal voluntary ventilation in 20 s (MVV₂₀), as well as ventilation and gas exchange variables during cycling were determined with an ergo-spirometric device, Oxycon Beta (Jaeger, Würzburg, Germany) using a turbine for volume measurements, a paramagnetic analyzer for O₂, and an infrared absorption analyzer for CO₂ measurements.

Maximal inspiratory mouth pressure (Pmax) was determined with a special device (Tecuria, Chur, Switzerland). This apparatus was also used for resistive breathing. It consists of a mouthpiece connected to a tube system including a flow sensor (163PC01D75, Honeywell, Phoenix, AZ) and a pressure sensor (143C05PCB, Sensym, Milpitas, CA). The tube system extends to two electronically controlled valves (inspiratory and expiratory). Breathing resistance increases proportionally to the voltage applied to the valves. Feedback on the generated mouth pressure is displayed on an oscilloscope. Cycling tests were performed on an electronically break cycle ergometer (Ergometrics 800 S, Ergoline, Bitz, Germany).

Core body temperature was measured by a rectal temperature probe (YSI Reusable Temperature Probe, Yellow Springs Instruments, Yellow Springs, OH) and monitored on a Duotemp TM101 (Fisher & Paykel, Auckland, New Zealand). Blood samples were drawn by a catheter inserted into a forearm vein. Blood lactate concentrations were determined enzymatically (Ebio 6666, Eppendorf, Hamburg, Germany), pH was measured with a blood gas analyzer (ILI304, Instrumentation Laboratory, Milano, Italy), and serum potassium concentrations were analyzed with a flame photometer (IL 943, Instrumentation Laboratory).

**Preliminary testing.** First, all subjects were familiarized with the different testing devices, in particular with the resistive breathing device as it is well known that subjects need to learn such a breathing technique (7). Spirometric measurements (VC, FEV₁, PEF, MVV₂₀) as well as Pmax maneuvers were performed until values were reproducible. Pmax was measured from residual volume (RV), whereas the subject performed a maximal inspiration against an occluded airway. An 18-gauge needle was inserted into the mouthpiece to ensure that the glottis stayed open (4,20). An incremental breathing test was then performed: subjects began by breathing against an inspiratory resistive load at a pressure corresponding to 60% Pmax with the exception of one subject who started at 55% Pmax, because he was not able to sustain a load of 60% Pmax for at least 3 min (see below). Expiration was unloaded and breathing frequency (fR) was set at 18 breath·min⁻¹ and paced by a metronome. Every 3 min, the resistive load was increased by 5% of Pmax. The test continued until the subjects were no longer able to overcome the load. The Pmax of the last step that the subjects were able to sustain for 3 min was selected as the target pressure for the constant-load test.

At least 2 d later, all subjects performed two consecutive resistive breathing tests at the predetermined, constant load (see above). The two tests were separated by a 15-min rest period. During each test, the subjects matched the mouth pressure to a pressure waveform (previously determined to be comfortable) displayed on the oscilloscope. All subjects breathed at an fR of 18 breath·min⁻¹. The maximal breathing endurance time was defined as the time when subjects were no longer able to overcome the load and/or to achieve the target pressure. During this test series, subjects were asked every minute to rate their respiratory exertion and air hunger on a modified Borg scale (24). This test series served as control to assure that the breathing tests that were performed after exercise in the main study (see below) would not be influenced by the baseline breathing test performed before exercise.

Study subjects performed an incremental cycling test to exhaustion to determine Wmax as well as VO₂peak. Starting at 100 W, the load was increased by 30 W every 2 min. The subjects chose their preferred pedaling frequency at the beginning of the test, and it was held constant thereafter. The highest load a subject could tolerate for at least 90 s was considered to be Wmax, the highest VO₂ measured over 15 s was determined to be VO₂peak.

**Main study.** The main tests were performed in random order on four different days separated by at least 48 h. Before each test, a catheter was inserted into the subjects’ forearm vein for blood sampling, and a rectal temperature probe was inserted and fixed with adhesive tape to prevent

<table>
<thead>
<tr>
<th>Group</th>
<th>VC (L)</th>
<th>FEV₁ (L)</th>
<th>PEF (L·s⁻¹)</th>
<th>MVV₂₀ (L·min⁻¹)</th>
<th>Pmax (cm H₂O)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Study</td>
<td>6.4 ± 0.6</td>
<td>4.8 ± 0.4</td>
<td>11.0 ± 1.0</td>
<td>194 ± 21</td>
<td>−165 ± 27</td>
</tr>
<tr>
<td>Control</td>
<td>5.9 ± 1.2</td>
<td>4.7 ± 0.8</td>
<td>11.3 ± 1.7</td>
<td>199 ± 36</td>
<td>−185 ± 28</td>
</tr>
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</table>

VC, vital capacity; FEV₁, forced expiratory volume in 1 s; PEF, peak expiratory flow; MVV₂₀, maximal voluntary ventilation in 20 s; Pmax, maximal inspiratory mouth pressure.
displacement. Before cycling ($t_0$), the subjects performed a constant-load breathing test to exhaustion to determine the maximal breathing endurance time. After a break of 15 min, subjects started cycling to exhaustion at either 65, 75, 85, or 95% VO$_{2peak}$. These cycling endurance tests were started at 100 W and, in order to reach the predetermined workload within 3 min, the workload was increased in three equal increments of 1 min duration. Ten ($t_{10}$) and 45 min ($t_{45}$) after the subjects stopped cycling, the breathing test to task failure was repeated. During each constant-load breathing test, subjects were asked to rate their respiratory exertion as well as their perception of air hunger on a modified Borg scale (24) every minute. Blood samples were drawn before and after each breathing test.

**Control series.** The subjects of the control group performed three constant load breathing tests separated by the same interval as the resistive breathing tests of the study group’s longest lasting test series (series with cycling at 65% VO$_{2peak}$). Also, subjects were asked to rate their respiratory exertion as well as their perception of air hunger on a modified Borg scale (24) every minute. This test series was performed to further assure that no decrease in respiratory endurance times measured after cycling in the main test series could possibly be a result of the preexercise resistive breathing test.

**Statistics.** An analysis of variance (ANOVA) with repeated measures was applied to compare variables of the constant-load breathing tests at $t_{10}$ and $t_{45}$ with variables at $t_0$ of the same test series, to compare the four constant-load breathing tests at the same time point ($t_0$, $t_{10}$ or $t_{45}$), and to compare cycling endurance times and ventilatory variables of the four different cycling endurance tests. If significance was found, Fisher’s PLSD post hoc test was used to locate the significant differences. Average values of blood lactate concentrations, pH, serum potassium concentrations, and core body temperature measured before and after the constant-load breathing tests were calculated for the above statistical comparisons. Variables of the two preliminary consecutive resistive breathing tests as well as baseline characteristics of the two groups were also compared by ANOVA. Results are given as mean ± SD. Values were considered to be significantly different if $P < 0.05$.

**RESULTS**

Breathing endurance was similarly reduced in constant-load breathing tests 10 min ($t_{10}$) after exhaustive cycling at 65, 75, 85, and 95% VO$_{2peak}$ as well as 45 min ($t_{45}$) after cycling at 65, 75, and 85% VO$_{2peak}$ (Fig. 1). The paced f$_R$ during the constant-load breathing tests at $t_0$, $t_{10}$, and $t_{45}$ were not significantly different between tests (average 18.4 ± 0.1 min$^{-1}$). Tidal volume (V$_T$) showed small but significant differences (Table 2). No significant differences were found in the ratings of perceived respiratory exertion or air hunger of the last minute of the constant-load breathing tests (Table 2).

The three breathing endurance tests of the control group were of similar length (first: 7.7 ± 4.4 min; second: 8.2 ± 4.1 min; third: 7.0 ± 3.3 min). The paced f$_R$ was the same in all three tests (18.2 ± 0.1 min$^{-1}$). Tidal volume did not differ significantly either (first: 1.11 ± 0.11 L; second: 1.06 ± 0.14 L; third: 1.06 ± 0.12 L). No significant differences were found in the ratings of perceived respiratory exertion (first: 8.3 ± 1.3; second: 9.0 ± 1.2; third: 9.2 ± 1.3) or air hunger (first: 7.8 ± 1.9; second: 9.0 ± 0.9; third: 8.5 ± 1.6) in the last minute of the constant-load breathing tests.

The exercise duration differed significantly among the cycling endurance tests (Table 3). Average power outputs during cycling at 65, 75, 85, and 95% VO$_{2peak}$ were 206 ± 24 W (67 ± 3% W$_{max}$), 238 ± 28 W (78 ± 2% W$_{max}$), 267 ± 32 W (87 ± 2% W$_{max}$), and 295 ± 37 W (96 ± 2% W$_{max}$). Steady-state ventilation (averaged ventilation during the constant load period of the cycling test excluding the last 2 min), total ventilation (sum of the ventilation over the entire cycling time), as well as ventilation during the last 2 min of each test (average ventilation of the last 2 min) were significantly different between tests (Table 3). Values for blood lactate concentrations (Fig. 2), pH (Fig. 3), serum potassium concentrations (Fig. 4), and core body temperature (Fig. 5) were not significantly different before the cycling endurance tests, but they differed significantly at $t_{10}$ after the cycling endurance tests.

Breathing endurance times of the two consecutive constant-load breathing tests were similar in the study group (6.6 ± 2.6 vs 6.9 ± 2.5 min) and in the control group (6.2 ± 2.2 vs 6.1 ± 2.3 min). Also, f$_R$ (study group: 18.4 ± 0.4 min$^{-1}$ vs 18.4 ± 0.4 min$^{-1}$; control group: 18.2 ± 0.1 min$^{-1}$ vs 18.2 ± 0.1 min$^{-1}$) was the same in both tests of both groups. Tidal volume was the same in both tests of the study group (0.82 ± 0.13 L vs 0.83 ± 0.15 L) while it was slightly smaller in the control group’s second test (1.14 ± 0.16 L vs 1.08 ± 0.18 L).
to exhaustion against a resistance, subjects’ motivation is crucial. To prevent lack of motivation influencing the outcome of the study, only highly motivated subjects were chosen to participate. In fact, ratings of perceived respiratory exertion and air hunger (Table 2) were similar at the end of all constant-load resistive breathing tests, suggesting that the subjects performed maximally.

Alternatively, a change in minute ventilation and/or breathing pattern during resistive breathing, as shown by Clanton et al. (5), could have been responsible for a reduced respiratory performance during tests at t10 and t45 compared with t0. In the present study, we did not observe any significant differences in fR between any of the breathing tests. However, mean VT during breathing tests at t45 after the 65%- and the 75%-cycling runs was slightly but significantly lower than preceeding VT. Thus, we assume that breathing endurance times would have been slightly smaller at t45 after the 65%- and 75%-cycling runs had VT been slightly higher, i.e., had VT been the same at t45 and at t0. A larger reduction in breathing endurance after 65%- and 75%-cycling runs than after 85%- and 95%-cycling runs would be even more surprising as we predicted less or no reduction in respiratory performance to occur after exhaustive cycling exercise at 65 and 75% VO2peak as the diaphragm hardly fatigues at these intensities (12,18).

Further, one could argue that a reduction of respiratory performance at t10 might be a consequence of preexisting fatigue of the respiratory muscles from breathing against the resistance at t0 as Laghi et al. (15) and Travaline et al. (23) have shown that diaphragmatic fatigue can last for at least 24 h when subjects breathe against inspiratory resistive loads of 60% of maximal Pdi (Pdi,max) for 33 min or 80% Pdi,max for 25 min. However, those loaded breathing tasks would be even more surprising as we predicted less or no reduction in respiratory performance to occur after exhaustive cycling exercise at 65 and 75% VO2peak as the diaphragm hardly fatigues at these intensities (12,18).

On the other hand, factors other than muscle fatigue may have led to the reduced respiratory muscle performance during constant-load resistive breathing after exhaustive cycling. In a subject-limited endurance test such as breathing

<table>
<thead>
<tr>
<th>Tidal volume</th>
<th>65% VO2peak</th>
<th>75% VO2peak</th>
<th>85% VO2peak</th>
<th>95% VO2peak</th>
</tr>
</thead>
<tbody>
<tr>
<td>at t0</td>
<td>0.88 ± 0.12</td>
<td>0.85 ± 0.11</td>
<td>0.85 ± 0.17</td>
<td>0.84 ± 0.11</td>
</tr>
<tr>
<td>at t10</td>
<td>0.80 ± 0.16</td>
<td>0.81 ± 0.19</td>
<td>0.87 ± 0.21</td>
<td>0.89 ± 0.16</td>
</tr>
<tr>
<td>at t45</td>
<td>0.78 ± 0.15*</td>
<td>0.76 ± 0.18*</td>
<td>0.80 ± 0.21</td>
<td>0.76 ± 0.17</td>
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<table>
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<tr>
<th>Perceived exertion</th>
<th>65% VO2peak</th>
<th>75% VO2peak</th>
<th>85% VO2peak</th>
<th>95% VO2peak</th>
</tr>
</thead>
<tbody>
<tr>
<td>at t0</td>
<td>9.4 ± 0.5</td>
<td>9.3 ± 0.8</td>
<td>9.7 ± 0.5</td>
<td>9.1 ± 1.1</td>
</tr>
<tr>
<td>at t10</td>
<td>8.9 ± 1.5</td>
<td>9.4 ± 1.0</td>
<td>9.4 ± 0.5</td>
<td>9.1 ± 1.1</td>
</tr>
<tr>
<td>at t45</td>
<td>9.4 ± 0.8</td>
<td>9.1 ± 0.9</td>
<td>8.7 ± 1.7</td>
<td>9.6 ± 0.5</td>
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<thead>
<tr>
<th>Air hunger</th>
<th>65% VO2peak</th>
<th>75% VO2peak</th>
<th>85% VO2peak</th>
<th>95% VO2peak</th>
</tr>
</thead>
<tbody>
<tr>
<td>at t0</td>
<td>8.9 ± 1.1</td>
<td>8.7 ± 1.1</td>
<td>8.9 ± 1.5</td>
<td>8.9 ± 1.3</td>
</tr>
<tr>
<td>at t10</td>
<td>8.8 ± 2.1</td>
<td>8.1 ± 2.2</td>
<td>8.1 ± 1.1</td>
<td>8.6 ± 1.0</td>
</tr>
<tr>
<td>at t45</td>
<td>8.9 ± 1.9</td>
<td>8.0 ± 1.3</td>
<td>7.4 ± 2.2</td>
<td>8.9 ± 1.1</td>
</tr>
</tbody>
</table>

* Significant differences of variables at t10 and t45 compared with t0 (P < 0.05).
the present study (average 9.0 ± 5.2 min). To assure that the recovery period between the preexercise breathing test and the start of cycling exercise would be long enough for the following breathing tests not to be affected by the first breathing test, all subjects performed a preliminary control test series. In this test series, respiratory muscle performance of the subjects was assessed in two subsequent constant-load resistive breathing tests to exhaustion with a 15-min pause in-between that previously proved to be long enough for a following breathing or cycling test not to be affected (21). Under these conditions, both breathing tests were of similar duration, suggesting that reduced breathing endurance times after cycling would not be caused by the preexercise breathing test. To also assure that resistive breathing tests dispersed over a period of almost 2 h (similar to the longest lasting series) would be of similar lengths without intervening cycling, an additional group of subjects was recruited. These subjects performed two test series: first they also completed the above described preliminary test series with two consecutive breathing tests with a 15-min-pause in-between that previously proved to be long enough for a following breathing or cycling test not to be affected

FIGURE 2—Mean blood lactate concentrations of the constant-load breathing tests (average of 10 subjects) before (t₀) as well as 10 (t₁₀) and 45 min (t₄₅) after cessation of an exhaustive cycling endurance test (CET) at 65, 75, 85, or 95% of peak oxygen uptake (V̇O₂peak). Significant differences of variables at t₁₀ and t₄₅ compared with t₀ are marked with asterisks (* P < 0.05; *** P < 0.001). Significant differences to values of the 65% series (**** P < 0.01; ***** P < 0.001), the 75% series (****** P < 0.001), and the 85% series (†† P < 0.05) compared at the same time point (t₀, t₁₀ or t₄₅) are also shown.

FIGURE 3—Mean blood pH of the constant-load breathing tests (average of 10 subjects) before (t₀) as well as 10 (t₁₀) and 45 min (t₄₅) after cessation of an exhaustive cycling endurance test (CET) at 65, 75, 85, or 95% of peak oxygen uptake (V̇O₂peak). Significant differences of variables at t₁₀ and t₄₅ compared with t₀ are marked with asterisks (**** P < 0.01; ***** P < 0.001). Significant differences to values of the 65% series (**** P < 0.01; ***** P < 0.001), the 75% series (****** P < 0.001), and the 85% series (†† P < 0.05) compared at the same time point (t₀, t₁₀ or t₄₅) are also shown.

FIGURE 4—Mean serum potassium concentrations of the constant-load breathing tests (average of 10 subjects) before (t₀) as well as 10 (t₁₀) and 45 min (t₄₅) after cessation of an exhaustive cycling endurance test (CET) at 65, 75, 85, or 95% of peak oxygen uptake (V̇O₂peak). Significant differences of variables at t₁₀ and t₄₅ compared with t₀ are marked with asterisks (* P < 0.05). Significant differences to values of the 65% series (°° P < 0.05) compared at the same time point (t₀, t₁₀ or t₄₅) are also shown.
pause, and then they performed three subsequent constant-load breathing tests to exhaustion without cycling in-between, the resting period between tests being similar to the time spans between breathing tests in the 65%-test series of the study group. As breathing endurance times of the three constant-load resistive breathing tests did not differ significantly in length in this control test series either, we suggest that the reduced breathing endurance times after cycling, at t₁₀, were a result of decreased respiratory performance due to the ventilatory work performed during exercise rather than being a result of fatigue from the precycling resistive breathing tests at t₀.

Finally, changes in blood lactate concentration, pH, serum potassium concentration, or core body temperature—factors known to influence muscle contractility (6,9,11) and performance (14)—might possibly account for the changes in respiratory performance after exhaustive cycling. Directly after cycling (t₁₀) at 85 and 95% VO₂peak, blood lactate concentrations were significantly higher and pH was significantly lower than after cycling at 65 and 75% VO₂peak. These changes would predict—if large enough to affect muscle contractility—a larger reduction in breathing endurance time after exercise at higher workloads. In contrast, serum potassium concentration and core body temperature were significantly higher at 65 than at 95% VO₂peak, which in turn would predict—if these changes were large enough to affect muscle contractility—a larger reduction in breathing endurance time after cycling at lower intensities. These different effects on muscle contractility do not need to be mutually exclusive and may in fact be additive. We could only speculate to which extent they possibly contributed to the decrease in respiratory muscle performance after exhaustive cycling but we believe that the changes were too small to have a major effect. Also, we believe that these small changes did not affect breathing endurance time, because most of these variables had reached baseline levels at t₄₅—but three of four breathing endurance times were still significantly reduced at this time.

To possibly explain why respiratory performance was reduced by similar degrees after exhaustive cycling at different intensities (65, 75, 85, and 95% VO₂peak), we compared the level of ventilation during the constant-load part of the cycling test as well as total ventilation summed over the entire exercise period as an index of total ventilatory work performed during the cycling test. As the level of steady-state ventilation was significantly lower during the 65% test, gradually increasing up to the test with the highest cycling workload, we can rule out the level of steady state ventilation as a possible reason for the similarly reduced respiratory performance after exercise. As the test performed at 65% VO₂peak lasted about 5 times longer than the 95%-test, one could argue that a lower ventilation held for a longer time might add up to the same ventilatory work as a larger ventilation performed over a shorter time and thus affecting respiratory muscles to a similar degree. We therefore summed ventilation over the entire exercise time: this total ventilation was significantly larger during the 65% test than during the 95% test, indicating that total ventilation per se cannot be responsible for the similarly decreased respiratory performance during all four breathing tests at t₁₀. Therefore, we additionally compared the ventilatory output during the last 2 min of exercise as it is known that only 2 min of maximal voluntary ventilation can cause respiratory muscle fatigue (8). Minute ventilation at the very end of the test may therefore be crucial for inducing respiratory muscle fatigue. As is evident from Table 3, there were significant differences of minute ventilation during these last minutes of exercise, ventilation being significantly smaller during the 65%-test than during the cycling tests at higher workloads, again indicating that the final ventilation per se cannot be responsible for the similar reduction in respiratory performance after different intensities of exercise.

Possibly a mixture of a long exercise time with a smaller ventilation and a smaller increase in ventilation at the end of exercise may result in the same impact on respiratory muscles as a short exercise time with a higher ventilation and a very high final output of the ventilatory system. To fully answer the seemingly contradicting results of previous studies and the present findings, further studies, possibly including ventilatory interventions and focusing on extra-diaphragmatic respiratory muscles during exercise, are needed.

**CONCLUSIONS**

The reduction of respiratory performance after exhaustive constant-load cycling tests at 65, 75, 85, and 95% VO₂peak was of similar degree when measured by exhaustive breathing against a constant resistive inspiratory load. These results contrast with measurements of diaphragmatic fatigue (reduced Pdi,tw), which is more pronounced after exercise of higher intensity. This difference possibly results from the involvement of extra-diaphragmatic muscles in addition to the diaphragm during resistive breathing.

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