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Inspiratory Muscles do not Limit Maximal Incremental Exercise Performance in Healthy Subjects

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Abstract

We investigated whether the inspiratory muscles affect maximal incremental exercise performance using a placebo-controlled, crossover design. Six cyclists each performed 6 incremental exercise tests. For 3 trials, subjects exercised with proportional assist ventilation (PAV). For the remaining 3 trials, subjects underwent sham respiratory muscle unloading (placebo). Inspiratory muscle pressure (P_{mus}) was reduced with PAV ($-35.9 \pm 2.3\%$ vs. placebo; $P < 0.05$). Furthermore, $\dot{V}_{O_2}$ and perceptions of dyspnea and limb discomfort at submaximal exercise intensities were significantly reduced with PAV. Peak workload, however, was not different between placebo and PAV (324 ± 4 vs. 326 ± 4 W; $P > 0.05$). Diaphragm fatigue (bilateral phrenic nerve stimulation) did not occur in placebo. In conclusion, substantially unloading the inspiratory muscles did not affect maximal incremental exercise performance. Therefore, our data do not support a role for either inspiratory muscle work or fatigue *per se* in the limitation of maximal incremental exercise performance.

Keywords

diaphragm; fatigue; respiratory muscles; dyspnea

1. Introduction

Exercise-induced diaphragm fatigue occurs in healthy individuals following heavy intensity (>85% maximal $\dot{V}_{O_2}$ uptake [$\dot{V}_{O_{2max}}$]), constant-load exercise performed to the limit of tolerance (Johnson et al. 1993; Babcock et al. 1998). There is mounting evidence that a metaboreflex effect from fatiguing respiratory muscles increases sympathetic outflow in the resting limb (St Croix et al. 2000; Derchak et al. 2002) and compromises perfusion of both

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resting (Sheel et al. 2001; Sheel et al. 2002) and exercising (Harms et al. 1997; Rodman et al. 2003) limb muscle, thereby limiting its ability to perform work during heavy exercise. When the inspiratory muscles are partially unloaded using a proportional assist ventilator (PAV), time-to-exhaustion during constant-load exercise is increased significantly (Harms et al. 2000) and diaphragm fatigue is prevented (Babcock et al. 2002).

It is unclear whether maximal incremental exercise elicits diaphragm fatigue in endurance-trained subjects (Levine et al. 1988; Verin et al. 2004). In addition, it is unknown whether partially unloading the inspiratory muscles during this type of exercise results in an increase in maximal power output. When the inspiratory muscles are mechanically unloaded during very heavy constant-load exercise, total $\dot{V}_{O_2}$ and cardiac output are reduced while leg blood flow and $\dot{V}_{O_2}$ are increased (Harms et al. 1997). We reason that such a redistribution of total blood flow and O_2 to the legs from the inspiratory muscles with unloading, secondary to a reduction in the severity of diaphragm fatigue, will increase the peak workload during maximal incremental exercise without a change in $\dot{V}_{O_{2max}}$. In the only study to partly investigate this postulate, Gallagher et al. (1989) did not find a significant effect of inspiratory muscle unloading via PAV on the duration of maximal incremental exercise. This previous study, however, did not quantify the magnitude of unloading. Indeed, $\dot{V}_{O_2}$ was not reduced with PAV at any time during exercise, which suggests that the inspiratory muscles were not unloaded optimally. Moreover, the study did not use a placebo group, which renders the findings difficult to interpret.

Thus, the purpose of the present study was to determine whether the inspiratory muscles limit maximal incremental exercise performance in endurance trained subjects. We hypothesized that: 1) diaphragm fatigue would occur after maximal incremental exercise; 2) mechanically unloading the inspiratory muscles via PAV would result in an increase in the peak power output achieved during incremental exercise; and 3) exercise-induced diaphragm fatigue would be prevented with PAV. Some of the results of this study have been reported previously in the form of an abstract (Romer et al. 2004).

2. Materials and Methods

2.1 Subjects

After receiving local ethics committee approval and written informed consent, 6 male cyclists with pulmonary function within normal limits participated in the study. Descriptive characteristics of the subjects (mean $\pm$ SEM) were: age 26.0 ± 2.3 y, stature 1.78 ± 0.04 m, body mass 73.8 ± 2.4 kg and $\dot{V}_{O_{2max}}$ 53.8 ± 1.1 ml·kg⁻¹·min⁻¹.

2.2 Study Design

The study used a placebo-controlled, crossover design. During at least two preliminary visits to the laboratory, subjects were familiarized with all of the procedures. During the first session, the subjects performed maximal incremental exercise and were familiarized with the phrenic nerve stimulation procedures. During this first session and during at least one further visit to the laboratory, the subjects practiced breathing on the ventilator. The remaining visits were used to assess maximal incremental exercise performance under experimental and placebo conditions. For the experimental condition, the subjects exercised

on 3 separate occasions while breathing on a proportional assist ventilator (PAV). For the placebo condition, the subjects exercised on 3 separate occasions while undergoing sham inspiratory muscle unloading. The order of trials was randomized and partially counterbalanced using a Latin square. The subjects were told that they were participating in a study to compare the effects of breathing on a mechanical ventilator (i.e., PAV) versus breathing a low-density gas mixture (i.e., placebo) on exercise performance and, as a consequence, were blinded to the true purpose of the study and the expected outcomes. Breathing mechanics and exercise-induced diaphragm fatigue were assessed in the final experimental and placebo trial by assessing spontaneous exercise pressure-volume data and transdiaphragmatic twitch pressures in response to bilateral phrenic nerve stimulation (BPNS), respectively. Each exercise session was separated by at least 48 h and was completed at the same time of day. Subjects refrained from caffeine for 12 h and stressful exercise for 48 h before each exercise test. Ambient temperature and relative humidity were not different between conditions.

2.3 Maximal Incremental Exercise

Maximal incremental exercise (600 kpm [98 W] + 200 kpm [33 W] every 3 min) was performed on an electromagnetically braked cycle ergometer (Elema, Sweden). Subjects selected their preferred pedaling cadence and the test was terminated when the cadence fell below 60 rev·min⁻¹ for more than 3 s. The subjects remained seated throughout exercise and received no information regarding either exercise intensity or time. Furthermore, the subjects received no verbal encouragement during the tests. Each subject's peak power output ($\dot{W}_{\text{peak}}$) was calculated as the sum of the workload in the last completed stage ($\dot{W}_{\text{lcs}}$) plus the fraction of time (t in seconds) in the final stage:

$$\dot{W}_{\text{peak}} = \dot{W}_{\text{lcs}} + (t/180) \cdot 35.$$

2.4 Inspiratory Muscle Unloading

A feedback controlled mechanical ventilator with a PAV mode was used to reduce the work of the inspiratory muscles during exercise (Younes 1992). During prior familiarization sessions, subjects were verbally coached to relax, reduce their inspiratory effort, and permit the ventilator to assist each inspiration as much as possible, as judged by the negativity of mouth pressure at the onset of each inspiration. The amount of assist was set at the maximum level each subject could tolerate (~ 2.5 cmH₂O·L·s⁻¹ for flow assist, ~ 3.0 cmH₂O·L⁻¹ for volume assist). These settings were chosen to cause decreases in inspiratory muscle work similar in magnitude to our previous studies, which had shown significant effects of changing inspiratory muscle work on diaphragm fatigue, limb blood flow, and performance during constant-load exercise (Harms et al. 1997; Harms et al. 2000; Babcock et al. 2002) (see Fig. 1 in online repository).

2.5 Sham Unloading

Before resting measurements were obtained and immediately after exercise had finished the subjects inspired briefly (~ 30 s) a HeO₂ gas mixture (fraction of inspired O₂ = 0.21) from a meteorological balloon. The resultant increase in voice pitch served to convince the subjects

that they were breathing HeO₂ throughout the entire exercise test. When the subjects went back on the mouthpiece the inspire was surreptitiously switched to room air.

2.6 Exercise Responses

Ventilatory and pulmonary gas exchange indices were measured breath-by-breath using apparatus and techniques that have been described previously (Johnson et al. 1992) (see also online repository). Arterial O₂ saturation was estimated (SpO₂) using a pulse oximeter (Nellcor N-595, Pleasanton, CA) with adhesive forehead sensors (Nellcor Max-Fast, Pleasanton, CA). Cardiac frequency (f_H) was measured from the R-R interval of an electrocardiogram (Hewlett Packard 7803B) using a three-lead arrangement. Ratings of perceived exertion (dyspnea and limb discomfort) were obtained at rest, in the final 30 s of each exercise stage, and for the end of exercise using Borg's modified CR10 scale (Borg 1998).

2.7 Breathing Mechanics

Gastric (Pg) and esophageal (Pe) pressures were measured using standard procedures (Baydur et al. 1982). Transdiaphragmatic pressure (Pdi) was obtained online by subtracting Pe from Pg. Transpulmonary pressure (P_{TP}) was calculated as the difference between mouth pressure (Pm) and Pe. Dynamic compliance of the lung (C_{l_{dyn}}) was calculated as the ratio between the change in V_T and the change in P_{TP} at zero flow points (Rodarte et al. 1986). From these measures of P_{TP} and C_{l_{dyn}}, end-expiratory lung volume (EELV) was estimated (Johnson et al. 1990). End-inspiratory lung volume (EILV) was calculated as the sum of EELV and V_T. Both EELV and EILV were expressed as a percentage of total lung capacity (TLC), as determined from whole-body plethysmography.

The flow resistive work performed on the lungs (W_L), defined as the integrated area of the Pe-V_T loop (Otis 1964), was calculated for every breath of the final minute of each incremental exercise stage. The calculated W_L per breath (cmH₂O · L⁻¹) was converted into joules per breath using a standard conversion factor (1 cmH₂O · L⁻¹ = 0.09806 J). The amount of work done per minute on the lungs (J · min⁻¹) was represented by W_L multiplied by respiratory frequency (f_R). The pressure produced by the inspiratory muscles on the respiratory system (P_{mus}) was estimated using an approach similar to that of Gallagher et al. (1989) (see online repository). Unlike Gallagher et al. (1989), we did not force P_{mus} to equal zero at the onset of inspiratory flow in the resting phase, and as such, our waveforms are shifted vertically. Although this approach may result in a slightly higher estimation of the absolute level of P_{mus}, this provides a more conservative measure of the relative reduction in P_{mus} during respiratory muscle unloading because the absolute decrease in P_{mus} with PAV would be unaffected.

To estimate the relative contributions of diaphragm and accessory inspiratory muscle contractions to the inspiratory volume excursion, Pdi and Pe were integrated over time during the period of inspiratory flow. These values, $\int Pdi \, dt$ and $\int Pe \, dt$, were multiplied by f_R to give an estimate of mean diaphragmatic pressure production and its ratio to total mean inspiratory muscle pressure production, respectively. One subject was unable to tolerate the

nasopharyngeal balloons; therefore, breathing mechanics data were collected for five subjects.

2.8 Diaphragm Fatigue

Two magnetic stimulators (Magstim 200, Jali Medical Inc., Newton, MA), connected to a transformer (TwinCap Module) and two custom-made double 40 mm coils, were used to stimulate the phrenic nerves from an anterolateral approach (Mills et al. 1996; Mador et al. 2002). We used single (nonpotentiated and potentiated) and paired stimuli (interstimulus intervals: 100 ms [10 Hz], 20 ms [50 Hz] and 10 ms [100 Hz]) to discriminate between low and high frequency fatigue (Yan et al. 1993; Polkey et al. 1997; Babcock et al. 1998). For paired stimuli the two stimulators were synchronized by a separate module (BiStim Module, Jali Medical Inc., Newton, MA). Membrane excitability, in response to nerve stimulation, was determined by measuring the peak-to-peak amplitudes of diaphragm compound muscle action potentials (M-waves) from skin surface electrodes (Luo et al. 2002). Evoked transdiaphragmatic twitch pressures ($P_{di,tw}$) were assessed at baseline, within 15 min post-exercise, and at 35 min post-exercise. At similar times, voluntary activation of the diaphragm during maximal Mueller maneuvers was assessed using a superimposed twitch technique (Bellemare et al. 1984). Briefly, the $P_{di,tw}$ produced during a maximal Mueller maneuver was compared to the potentiated $P_{di,tw}$ produced ~5 s afterwards. Specific methodological details regarding the assessment of diaphragm fatigue are included in the online repository.

2.9 Data Analyses

Two-way repeated measures ANOVA (time and condition) was used to test for within-group effects for each variable. The reliability of test measures was expressed as the mean coefficient of variation (CV) for each subject. The slope of the relationship between Borg scale indices (dyspnea and limb discomfort) and power output was calculated for each data set using an equation of the form $y = ax^2$, where 'y' is the Borg score (arbitrary units), 'x' is power output (in W), and 'a' is the curvature constant. The origin of this parabolic function was 'forced' through the resting value. Curves were fit to the Borg score data using the 8.2 release version of SAS (SAS Institute Inc, Cary, NC). Results are expressed as the mean $\pm$ standard error of the mean (SEM), unless stated otherwise. Statistical significance was set at $P < 0.05$. Statistical analyses were performed using SPSS 11.5 (SPSS Inc., Chicago, IL).

3. Results

3.1 Inspiratory Muscle Unloading

The flow resistive work performed on the lungs (W_L) and the pressure produced by the inspiratory muscles on the respiratory system (P_{mus}) were significantly reduced during PAV versus placebo across all workloads (mean $-55.3 \pm 7.0\%$ and $-35.9 \pm 2.3\%$, respectively; Table 1). At every workload, including the final minute of exercise, oxygen uptake ($\dot{V}_{O_2}$) was significantly less with PAV vs. placebo (mean across all stages $-6.5 \pm 0.2\%$) (Table 1).

Group mean force output of the diaphragm ($\int \text{Pdi} \cdot f_R$), and the total inspiratory muscle force output of the diaphragm and accessory inspiratory muscles ($\int \text{Pe} \cdot f_R$) are shown in Fig. 1. During placebo, $\int \text{Pdi} \cdot f_R$ increased sharply from rest to $\sim 50\% \dot{W}_{\text{max}}$ and continued to increase at a slower rate thereafter. In contrast, $\int \text{Pdi} \cdot f_R$ during PAV increased by only 12% of the resting value and leveled off thereafter. The $\int \text{Pe} \cdot f_R$ increased progressively throughout exercise, although the increase from rest was less for PAV ($\sim 99\%$) than for placebo ($\sim 170\%$). The relative contribution of the diaphragm to the total inspiratory muscle force output ($\int \text{Pdi} / \int \text{Pe}$) during inspiration was gradually reduced to $\sim 65\%$ by the end of exercise, although there was no difference between placebo and PAV.

3.2 Incremental Exercise Performance

There was no difference ($P = 0.63$; 95% CI = -4.8 to 5.4 W) in $\dot{W}_{\text{peak}}$ between placebo (324.2 ± 4.2 W) and PAV (326.0 ± 3.7 W). Total exercise time was also not different ($P = 0.63$) between placebo (23.6 ± 0.4 min) and PAV (23.8 ± 0.3 min). Individual subject data are presented in the online repository (Fig. 3). The group mean within subject, between-day reliability of $\dot{W}_{\text{peak}}$ was similar for placebo (CV = $2.9 \pm 1.0\%$) and PAV (CV = $2.7 \pm 0.4\%$).

3.3 Exercise Responses

Group mean data for the physiological responses to exercise are shown in Table 1. None of the subjects showed signs of exercise-induced arterial hypoxemia ($\text{SpO}_2 > 95\%$). An increase in tidal volume (V_T) and a tendency for respiratory frequency (f_R) to be lower with unloading resulted in values for minute ventilation ($\dot{V}_E$) that were not different between placebo and PAV at any exercise intensity. The increase in V_T with PAV versus placebo, coupled with a trend toward a reduction in inspiratory time (t_i) resulted in a slight, albeit nonsignificant, increase in mean inspiratory flow ($\dot{V}_T/t_i$) with PAV across all exercise stages. At the onset of exercise end-expiratory lung volume (EELV) was reduced below resting values during both placebo and PAV trials, and then rose throughout the rest of exercise to values greater than FRC. For both placebo and PAV, the end-inspiratory lung volume (EILV) increased progressively throughout exercise.

The perceptual responses to exercise are shown in Fig. 2. Borg dyspnea ratings during the middle stages of exercise (50 to 80% $\dot{W}_{\text{peak}}$) were reduced with PAV relative to placebo ($P = 0.014$ to 0.034). Furthermore, ratings of limb discomfort were lower during the later stages of exercise (70 to 90% $\dot{W}_{\text{peak}}$) with PAV versus placebo ($P = 0.018$ to 0.024). There was no difference in either dyspnea or limb discomfort between placebo and PAV at end exercise. A parabolic function provided excellent fit to the Borg score/power-output data ($R^2 = 0.971 \pm 0.005$ for dyspnea and 0.984 ± 0.002 for limb discomfort). The curvature constant of the parabolic RPE (dyspnea)/power-output relationship was not different between PAV ($676 \times 10^7 \pm 57 \times 10^7$) and placebo ($838 \times 10^7 \pm 50 \times 10^7$) ($P = 0.059$). However, the curvature constant of the RPE (limb discomfort)/power-output relationship was significantly lower ($P = 0.024$) with PAV ($819 \times 10^7 \pm 41 \times 10^7$) vs. placebo ($896 \times 10^7 \pm 38 \times 10^7$).

3.4 Diaphragm Fatigue

Membrane excitability was not affected by the exercise, as evidenced by nonsignificant changes in diaphragm EMG M-wave amplitudes pre- vs. post-exercise. For placebo, there was a post-exercise reduction in $P_{di,tw}$ at 50 and 100 Hz ($P = 0.013$ and 0.023 , respectively; Fig. 3). The single potentiated $P_{di,tw}$ was not different pre- vs. post-exercise (56.7 ± 5.8 vs. 50.2 ± 4.3 cmH₂O at <15 min post-exercise; $P = 0.051$). Within-twitch responses for the 1 Hz stimulus did not differ pre- vs. post-exercise. For the maximal Mueller maneuvers there were nonsignificant changes pre- vs. <15 min post-exercise in $P_{m,max}$ (-119 ± 9 vs. -120 ± 12 cmH₂O; $P = 0.85$), $P_{di,max}$ (138 ± 16 vs. 131 ± 15 cmH₂O; $P = 0.31$), and voluntary activation (85 ± 6 vs. $83 \pm 8\%$; $P = 0.35$).

4. Discussion

4.1 Main Findings

We are the first to investigate specifically whether the work of breathing affects maximal incremental exercise performance. Despite substantial unloading of the inspiratory muscles, plus significant reductions in whole-body O₂ uptake ($\dot{V}_{O_2}$) and the perceptual responses to exercise (dyspnea and limb discomfort), maximal incremental exercise performance was unchanged with PAV versus sham unloading (placebo). Furthermore, using supramaximal bilateral phrenic nerve stimulation (BPNS) we found no clear evidence of exercise-induced diaphragm fatigue. These findings suggest that under the conditions of the present study, neither inspiratory muscle work nor diaphragm fatigue limit maximal incremental exercise performance in moderately-fit subjects. Whether there is an effect in more highly-trained subjects is unknown. However, given that the work of breathing during near-maximal exercise requires ~10% of total $\dot{V}_{O_2}$ for moderately trained subjects compared with ~15% in highly fit subjects at higher peak work rates and pulmonary ventilation (Aaron et al. 1992), it is reasonable to presume that the effect of reducing the work of breathing during maximal incremental exercise would be most noticeable in the highly fit subject.

4.2 Comparison with Previous Studies

4.2.1 Exercise performance—Our finding of a non-significant effect of inspiratory muscle unloading via PAV on maximal incremental exercise performance is in agreement with Gallagher et al. (1989). Although the primary aim of this previous study was to determine the effect of PAV on ventilation and respiratory mechanics, a nonsignificant difference in the duration of maximal incremental cycle exercise (50 W every 3 min, total exercise time of 16 min) was noted between control and PAV. These authors did not measure the magnitude of unloading during incremental exercise, but a significant reduction (21%) in mean inspiratory P_{mus} during *constant load* exercise (~70-80% of $\dot{V}_{O_{2max}}$) was reported when mouth pressure during inspiration was made positive and proportional to flow. By comparison, we found even larger reductions in mean inspiratory P_{mus} (36% at ~71% of $\dot{V}_{O_{2max}}$) by using a combination of flow and volume assist. These larger reductions in P_{mus} might explain why we also found significant reductions in $\dot{V}_{O_2}$ throughout incremental exercise whereas Gallagher and colleagues did not. Further advantages of our study are that we used several practice trials and multiple, randomized performance trials in an attempt to reduce the random variability of $\dot{W}_{peak}$. Indeed, the

between-occasion reliability of $\dot{V}_{peak}$ was excellent (CV <3%) and similar to that reported previously using a similar protocol in fit subjects (CV 3-6%) (Moseley et al. 2001). Importantly, our study incorporated a placebo group in the experimental design whereby sham unloading was used to overcome the subject expectation associated with PAV. Using this approach, subjects were deceived into thinking that they were breathing a gas mixture that would reduce the work of breathing and potentially improve exercise performance. Studies without a placebo group (Gallagher et al. 1989) can be criticized for having weak internal validity and for being vulnerable to the potential influence of subject bias.

4.2.2 Inspiratory muscle fatigue—Exercise-induced inspiratory muscle fatigue has been defined as a decrease in the capacity for developing inspiratory muscle pressure, resulting from whole body exercise, and which is reversible by rest (NHLBI-Workshop 1990). Across a wide range of stimulus frequencies (1-100 Hz), transdiaphragmatic twitch pressure in response to BPNS was not different at any time post- vs. pre-exercise. Furthermore, the within-twitch contractile properties were unchanged after exercise. Thus, it appears that the diaphragm does not fatigue in response to maximal incremental exercise in young, moderately-fit male subjects. This result is in agreement with previous studies in healthy, albeit untrained subjects (Levine et al. 1988; Verin et al. 2004). Levine et al. (1988) used BPNS to show that diaphragm fatigue occurred after maximal incremental treadmill exercise in approximately 50% of subjects, but only when an inspiratory resistance was added to the breathing circuit. More recently, Verin et al. (2004) found, also using BPNS, that maximal incremental treadmill exercise did not elicit significant peripheral fatigue of the diaphragm in healthy subjects.

In addition to the non-significant change in evoked diaphragm pressures, maximal volitional inspiratory mouth pressure ($P_{m,max}$) was not different after exercise compared with baseline values. As $P_{m,max}$ is a measure of the combined strength of the diaphragm and accessory inspiratory muscles, it is reasonable to conclude that global inspiratory muscle fatigue did not occur in the present study. A significant reduction in $P_{m,max}$ in response to maximal incremental exercise has been reported by some (Coast et al. 1990; McConnell et al. 1997) but not all previous studies (Younes et al. 1984; Coast et al. 1990). Between-study differences in the fitness level of subjects, the duration of the exercise test, and the timing of post-exercise measurements may have contributed, at least in part, to the discrepancies. Nevertheless, a limitation of using maximal volitional inspiratory maneuvers to quantify the magnitude of inspiratory muscle fatigue is that it is impossible to determine whether the fatigue is central or peripheral in origin. In the present study we used the twitch-interpolation technique to examine for central fatigue. Using this technique we found no evidence of exercise-induced central activation failure of the diaphragm. This finding is in agreement with one previous study (Gudjonsdottir et al. 2001), although more recent evidence using transcranial magnetic stimulation suggests that central “supraspinal” diaphragm fatigue may occur in response to maximal incremental exercise (Verin et al. 2004).

4.3 Why Did Inspiratory Muscle Unloading Not Affect Maximal Incremental Exercise Performance?

We hypothesized that reducing the normally occurring work of breathing with PAV would increase the peak power output achieved during maximal incremental exercise via effects on O₂ transport, inspiratory muscle fatigue, and the perception of respiratory and/or locomotor muscle effort.

4.3.1 O₂ transport to locomotor muscles—Based on our previous findings (Harms et al. 1997; Harms et al. 1998; Wetter et al. 1999) we predict that, toward the higher intensities of incremental exercise, limb vascular conductance, blood flow and O₂ transport increased with inspiratory muscle unloading. We hypothesized that such a redistribution of total blood flow and O₂ to the legs from the respiratory muscles would increase the power output achieved during maximal incremental exercise. It is important to emphasize, however, that mechanical ventilation creates a less negative intrathoracic pressure that, via a reduction in venous return, reduces stroke volume and cardiac output (Harms et al. 1998). Thus, the effects of PAV on limb blood flow and limb V_{O₂}, although consistent and significant, are relatively small (~5-7% above control) (Harms et al. 1997). Nevertheless, limb muscle force output is responsive to even small changes in limb muscle blood flow during high intensity muscle contractions (Barclay 1986). Furthermore, we have recently shown that the limb muscle fatigue elicited by sustained high-intensity exercise (>90% of V_{O_{2max}}) is attenuated when the inspiratory muscles are partially unloaded to a level similar to that achieved in the present study (Romer et al. 2006). That $\dot{W}_{\max}$ was unaffected by PAV suggests either that limb fatigue was unaffected by inspiratory muscle unloading or that exercise tolerance during maximal incremental exercise is unaffected by such changes in limb fatigue.

4.3.2 Inspiratory muscle fatigue—The inconsistent decreases in transdiaphragmatic twitch pressure after maximal incremental exercise (–15% across stimulus frequencies) were less than the reductions typically observed after sustained, high-intensity exercise (–20 to 32%) (Johnson et al. 1993; Babcock et al. 1998). That maximal incremental exercise did not elicit significant diaphragm fatigue points to the critical importance of exercise duration *and* intensity in defining the fatigability of the diaphragm and its role in exercise performance. Consistent reductions in transdiaphragmatic twitch pressures occur only at exercise intensities eliciting 90-95% of V_{O_{2max}} whereby exercise durations are at least 8-10 min (Johnson et al. 1993). In the present study, subjects exercised above 90% of V_{O_{2max}} for only 4.3 min, which was possibly not enough time for the cumulative work history of the diaphragm to reach fatiguing levels.

Despite limited evidence of diaphragm fatigue in response to maximal incremental exercise, we have recently shown that the limb locomotor muscles of endurance-trained cyclists (n = 12) exhibit significant peripheral and central fatigue in response to such exercise (Romer et al. 2005). Thus, using the same exercise and stimulation protocols as in the present study we found a significant depression of quadriceps twitch force across a wide range of stimulus frequencies (mean –33%, 1-100 Hz) that was recoverable only in part by 70 min of rest. In addition, there was a significant reduction in voluntary activation (–9%), as assessed via twitch interpolation, immediately after the exercise. These findings, in combination with

those from the present study, suggest that task failure during incremental exercise may be linked more to locomotor muscle fatigue than to fatigue of the diaphragm.

4.3.3 Perceptual ratings—Mechanical unloading of the inspiratory muscles with PAV resulted in significant reductions in the absolute ratings of dyspnea and limb discomfort throughout the later stages of incremental exercise. Furthermore, there was a trend for the “slope” of the RPE/power output relationships to be lower with PAV versus placebo, although statistical significance was only achieved for limb discomfort. Although no other study has reported the effect of mechanical unloading on the perceptual responses to incremental exercise, several studies have shown that perceptual ratings are reduced during constant load exercise (Marciniuk et al. 1994; Krishnan et al. 1996; Harms et al. 2000).

The reduction in dyspnea with inspiratory muscle unloading was likely due to a decrease in the level of motor outflow to these muscles resulting from a decrease in the developed inspiratory muscle tension (ATS 1999). Although inspiratory muscle fatigue and/or changes in respiratory muscle recruitment may also affect the severity of dyspnea (Bradley et al. 1986; Supinski et al. 1987), these factors did not change appreciably with unloading and, therefore, are unlikely to account for the changes noted in the present study. In addition to the effects of respiratory muscle unloading on dyspnea, PAV also mediated the perception of peripheral effort. We hypothesized that PAV would lower the relative intensity of inspiratory muscle work (Sheel et al. 2002) and/or delay the fatigue of the inspiratory muscles (Babcock et al. 2002), resulting in the production of fewer metabolic stimuli by these muscles. A reduction in chemical stimuli from respiratory muscles would be expected to reduce the sympathetic outflow to the working limbs via type III/IV afferents, thereby reducing vasoconstriction in the limbs (Harms et al. 1997; Sheel et al. 2001; Rodman et al. 2003). The resultant increase in limb blood flow would increase O₂ delivery to the limbs (Harms et al. 1997), reduce limb fatigue (Romer et al. 2006), and attenuate peripheral effort sensations (Harms et al. 2000; Romer et al. 2006). That exercise-induced inspiratory muscle fatigue was not a consistent finding in the present study suggests that redistribution of cardiac output to the exercising limb vasculature did not contribute significantly toward the reduction in limb discomfort with unloading. An alternative explanation for why the perception of limb discomfort was reduced with unloading is that subjects may have been unable to discriminate with much sensitivity between these two noxious sensations.

Although PAV relieved dyspnea and limb discomfort during the later stages of incremental exercise, there were no differences between placebo and PAV towards the end of exercise. At the high airflows and lung volumes achieved toward the end of maximal incremental exercise the pressures delivered by the ventilator were substantial (~30 cmH₂O peak inspiratory mouth pressure – see Fig. 2 in online repository). It is possible that these high pressures were disruptive to the subjects, which may explain in part why perceptual ratings were not different at the end of exercise even though the work of breathing was lower with PAV throughout exercise. In turn, the subjects' attention may have been diverted from the main task of exercising to maximum, which might have contributed to the non-significant change in exercise performance with inspiratory muscle unloading.

4.4 Conclusions

Significant inspiratory muscle unloading, which resulted in reductions in both whole body $\dot{V}_{O_2}$ and the perceptual responses to exercise, did not significantly affect maximal incremental exercise performance when comparisons were made with results from a sham unloading protocol. In addition, there was limited evidence for either central fatigue or peripheral inspiratory muscle fatigue under the conditions of the present study. Consequently, our data do not support the notion that the inspiratory muscles compromise exercise performance during maximal incremental exercise in moderately-fit subjects. Whether the inspiratory muscles limit maximal incremental exercise in highly fit subjects remains unknown.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Fig. 1.

Group mean ($n = 5$ subjects) diaphragm force output ($\int P_{di} dt \cdot f_R$; top panel), total inspiratory muscle force output ($\int P_e dt \cdot f_R$; middle panel), and the relative contribution of the diaphragm to the total inspiratory muscle force output ($\int P_{di} / \int P_e$; bottom panel) for placebo (closed circles) and PAV (open circles). ** $P < 0.01$, significantly different vs. placebo.

Fig. 2.

Group mean data ($n = 6$ subjects) for ratings of dyspnea (top panel) and limb discomfort (bottom panel) versus exercise intensity for placebo (closed circles) and PAV (open circles).

* $P < 0.05$, significantly different vs. placebo.

Fig. 3.

Individual and group mean (n = 5 subjects) transdiaphragmatic twitch pressure (Pdi,tw) response to supramaximal bilateral phrenic nerve stimulation at 1 Hz (nonpotentiated), 10 Hz, 50 Hz, and 100 Hz before, within 15 min and at 35 min after maximal incremental exercise for placebo condition. * $P < 0.05$, significantly different vs. pre-exercise (Pre).

Table 1

Physiological responses to incremental exercise for the placebo and the PAV groups.

	131 W		229 W		314 ± 8 W	
	Placebo	PAV	Placebo	PAV	Placebo	PAV
$\dot{V}_{Pe} dV$, J · min ⁻¹	74.1 ± 9.3	30.7 ± 6.1**	149.0 ± 18.5	53.5 ± 9.9**	342.3 ± 25.5	164.6 ± 37.0**
$\dot{V}_{Pmus} dV$, cmH ₂ O · L ⁻¹	62.7 ± 7.8	43.9 ± 4.8**	94.7 ± 5.5	71.4 ± 3.5**	150.8 ± 9.2	122.9 ± 6.9**
SpO ₂ , %	99.0 ± 0.3	99.6 ± 0.2	96.5 ± 0.3	97.4 ± 0.3	95.6 ± 0.3	96.4 ± 0.3
f _{ti} , beats·min ⁻¹	119 ± 3	114 ± 3	152 ± 3	148 ± 4	179 ± 2	176 ± 4
f _r , breaths·min ⁻¹	23.7 ± 1.1	21.6 ± 1.1	31.4 ± 1.2	28.0 ± 1.2	49.1 ± 1.1	44.4 ± 1.8
V _T , L	2.45 ± 0.06	2.77 ± 0.12	2.92 ± 0.07	3.37 ± 0.13*	3.14 ± 0.09	3.63 ± 0.14*
$\dot{V}_{E}$, L·min ⁻¹	56.5 ± 1.7	58.1 ± 1.2	90.0 ± 2.2	92.8 ± 2.1	152.4 ± 3.8	159.9 ± 5.8
ti/tot	0.443 ± 0.006	0.430 ± 0.014	0.454 ± 0.003	0.424 ± 0.011	0.472 ± 0.005	0.419 ± 0.009*
V _T /ti, L·s ⁻¹	2.14 ± 0.06	2.30 ± 0.08	3.31 ± 0.07	3.66 ± 0.11	5.39 ± 0.12	6.37 ± 0.08
EELV, %TLC	38.8 ± 3.1	35.3 ± 3.2	43.8 ± 4.9	39.5 ± 3.9	59.0 ± 6.0	49.0 ± 4.1
EILV, %TLC	73.0 ± 2.4	74.0 ± 2.8	84.0 ± 2.9	86.6 ± 2.5	100.0 ± 3.5	99.5 ± 1.8
$\dot{V}_{O_2}$, L·min ⁻¹	2.06 ± 0.04	1.96 ± 0.04*	3.09 ± 0.04	2.96 ± 0.03*	3.97 ± 0.08	3.72 ± 0.04*
$\dot{V}_{CO_2}$, L·min ⁻¹	1.90 ± 0.04	1.78 ± 0.04*	3.12 ± 0.06	3.03 ± 0.05*	4.35 ± 0.10	4.19 ± 0.06*
RER	0.92 ± 0.01	0.91 ± 0.01	1.01 ± 0.01	1.02 ± 0.01	1.10 ± 0.01	1.13 ± 0.02
P _{ETCO₂} , mmHg	43.2 ± 0.7	41.5 ± 0.9	41.6 ± 0.9	42.8 ± 0.6	33.2 ± 0.8	34.1 ± 0.8
RPE (dyspnea)	1.2 ± 0.1	1.1 ± 0.2	4.3 ± 0.3	3.3 ± 0.3*	9.1 ± 0.2	8.4 ± 0.4
RPE (limb)	1.2 ± 0.1	1.1 ± 0.1	4.6 ± 0.3	3.8 ± 0.3*	9.7 ± 0.2	9.5 ± 0.2

Values are mean ± SEM for stage 2 (131 W), stage 5 (229 W) and the final completed stage (314 ± 8 W) of incremental exercise. $\dot{V}_{Pe} dV$, volume integrated inspiratory esophageal pressure; $\dot{V}_{Pmus} dV$, volume integrated inspiratory muscle pressure; SpO₂, arterial O₂ saturation; f_{ti}, cardiac frequency; f_r, respiratory frequency; V_T, tidal volume; $\dot{V}_{E}$, minute ventilation; ti/tot, inspiratory duty cycle; V_T/ti, mean inspiratory flow; EELV, end-expiratory lung volume expressed as a percentage of total lung capacity; EILV, end-inspiratory lung volume expressed as a percentage of total lung capacity; $\dot{V}_{O_2}$, O₂ uptake; $\dot{V}_{CO_2}$, CO₂ production; RER, respiratory exchange ratio; P_{ETCO₂}, partial pressure of end-tidal CO₂; RPE, ratings of perceived exertion. n = 6 subjects.

* $P < 0.05$;

** $P < 0.01$, significantly different vs. placebo.