

Title:

1 Training the inspiratory muscles improves running performance when carrying a 25 kg
2 thoracic load in a backpack.

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12 Load Carriage and Inspiratory Muscle Training

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20

Abstract

1 Load carriage exercise in physically demanding occupations is typically characterised by
2 periods of low intensity steady state exercise and short duration, high-intensity exercise
3 whilst carrying an external mass in a backpack; this form of exercise is also known as load
4 carriage exercise (LC). This induces inspiratory muscle fatigue and reduces whole-body
5 performance. Accordingly we investigated the effect of inspiratory muscle training (IMT,
6 50% maximal inspiratory muscle pressure [$P_{I_{max}}$] twice daily for 6 wk) upon running time
7 trial performance with thoracic load carriage. Nineteen healthy males formed a pressure
8 threshold IMT ($n=10$) or placebo control group (PLA; $n=9$) and performed 60 min load
9 carriage exercise (LC, $6.5 \text{ km}\cdot\text{h}^{-1}$) followed by a 2.4 km running time trial (LC_{TT}) either side
10 of a double-blind six week intervention. Prior to the intervention, $P_{I_{max}}$ was reduced relative
11 to baseline, post-LC and post LC_{TT} in both groups (pooled data: $13 \pm 7\%$ and $16 \pm 8\%$,
12 respectively, $p<0.05$) and similar changes were observed post-PLA. Post IMT only, resting
13 $P_{I_{max}}$ increased $+31\%$ ($p<0.05$) and relative to pre-IMT was greater post-LC ($+19\%$) and
14 post-LC_{TT} ($+18\%$, $p<0.05$), however, the relative reduction in $P_{I_{max}}$ at each time point was
15 unchanged ($13 \pm 11\%$ and $17 \pm 9\%$, respectively, $p>0.05$). In IMT only, heart rate and
16 perceptual responses were reduced post-LC ($p<0.05$). Time trial performance was unchanged
17 post-PLA and improved $8 \pm 4\%$ after IMT ($p<0.05$). In summary, when wearing a 25 kg
18 backpack, IMT attenuated the cardiovascular and perceptual responses to steady state
19 exercise and improved high-intensity time trial performance which we attribute in part to
20 reduced relative work intensity of the inspiratory muscles due to improved inspiratory muscle
21 strength. These findings have real-world implications for occupational contexts.

Key Words

- 1 Inspiratory muscle training, fatigue, load carriage, performance

Introduction

1 Thoracic load carriage is defined as locomotion while bearing a mass upon the torso
2 supported by shoulder straps and / or a hip belt (i.e. a backpack) (Knapik, Harman, Steelman,
3 & Graham, 2012) and is common in physically demanding occupations (e.g. deployed
4 military and emergency services) and recreational groups. Carrying a backpack remains a
5 convenient and economical way of transporting external loads, particularly in some military
6 contexts where land vehicles may be restricted (Knapik et al., 2012). The placement of the
7 load upon the thorax, causes a volume limitation which increases the work of breathing
8 (Dominelli, Sheel, & Foster, 2012; Faghy & Brown, 2014b), this places the respiratory
9 musculature outside the optimal boundaries of their length-tension curve (Romer &
10 McConnell, 2004). The consequence of such a change in breathing mechanics is respiratory
11 muscle fatigue (Faghy & Brown, 2014a, 2014b) which may impair whole-body performance
12 by attenuating locomotor muscle blood flow and heightening perception of effort (Dempsey,
13 Romer, Rodman, Miller, & Smith, 2006; Harms et al., 1997).

14 Inspiratory muscle fatigue has been documented after constant power, high intensity, short
15 duration exercise (10 to 15 min) to the limit of volitional tolerance (~85% $\dot{V}O_2$ peak)
16 (Dempsey et al., 2006), high-intensity time trial exercise (Johnson, Sharpe, & Brown, 2007;
17 Romer, McConnell, & Jones, 2002b) and prolonged (120 to 180 min) submaximal exercise
18 such as marathon running (Ross, Middleton, Shave, George, & McConnell, 2008). In an
19 occupational context however, there is a dearth of literature exploring the consequences of
20 thoracic loads upon respiratory muscle function. Previous research has observed reductions in
21 maximum respiratory pressures following sub-maximal and intense treadmill exercise while
22 wearing self-contained breathing apparatus (SCBA) and fire-fighter apparel (Butcher, Jones,
23 Eves, & Petersen, 2006). However, SCBA incorporates a gas regulator (similar to those worn
24 by firefighters), delivering high-pressure air (~5 cmH₂O) from compressed-air cylinders and

1 it is difficult to distinguish the effects of the load mass secured to the thorax (from the gas
2 cylinders) and the positive pressure provided by the face mask since the latter accounts for
3 the majority of the increased work of breathing experienced during exercise (Eves, Jones, &
4 Petersen, 2005). Exercise in physically demanding occupations comprises low intensity
5 steady state exercise (e.g., during military patrol) and high intensity exercise (e.g., during
6 military engagement). In a military context, carrying a 25kg thoracic load we previously
7 demonstrated an 11% reduction in $P_{I_{max}}$ following moderate duration (60 min) low intensity
8 treadmill walking (~58% $\dot{V}O_{2peak}$) which further decreased 5% following a high-intensity
9 2.4 km self-paced time-trial (Faghy & Brown, 2014a, 2014b).

10 Inspiratory muscle training (IMT) has been employed to reduce inspiratory muscle fatigue
11 (for a review see HajGhanbari et al., 2013; Illi, Held, Frank, & Spengler, 2012). IMT
12 increases the strength of the chest wall inspiratory muscles and the diaphragm (P. I. Brown,
13 Johnson, & Sharpe, 2014) with exercise-induced inspiratory muscle fatigue either attenuated
14 (Romer & McConnell, 2004) or unchanged (Verges, Lenherr, Haner, Schulz, & Spengler,
15 2007). IMT also improves whole-body time-trial performance in cycling (Johnson et al.,
16 2007), running (Tong et al., 2008) and rowing (Griffiths & McConnell, 2007). The effect of
17 IMT upon inspiratory muscle fatigue following exercise with load carriage and the effects of
18 this upon load carriage performance remains unknown. Accordingly the aim here was to
19 investigate the effects of 6 wk IMT upon physiological and perceptual responses during 60
20 min submaximal walking exercise and upon 2.4 km self-paced running time-trial while
21 carrying a 25kg load. We hypothesised that IMT would increase inspiratory muscle strength;
22 reduce inspiratory muscle fatigue and in turn, improve time trial performance.

Methods

1 Participants

2 Following ethics approval from the host University and written informed consent, 19
3 healthy, non-smoking and physically active males fully habituated with load carriage through
4 regular recreational load carriage activities completed a double-blind placebo controlled
5 intervention. Blinding of the participants and principal investigators to the training conditions
6 was completed by an independent technician. Participants were separated in to an IMT group
7 ($n=10$) or a placebo control group (PLA; $n=9$) with no between group differences in any
8 baseline measurements (Table 1, $P>0.05$). Participants were instructed to avoid strenuous
9 exercise on the day preceding and the day of each exercise test. Participants abstained from
10 alcohol and caffeine and completed a 24h diet record prior to their first preliminary trial, to
11 ensure absence of alcohol and caffeine and also to ensure that nutritional intake was identical
12 in the 24 hours preceding each trial.

13 *Experimental Design*

14 *Preliminary Trials*

15 Participants completed two preliminary trials and the experimental trial each separated by
16 a minimum of seven days. First, body composition was assessed using dual energy x-ray
17 absorptiometry (Lunar iDXA, GE Healthcare, Hertfordshire, UK) followed by a maximal
18 incremental exercise test to determine peak O₂ uptake ($\dot{V}O_{2peak}$) performed on a motorised
19 treadmill (Desmo, Woodway, Germany). Following a 5 min warm-up at 8 km·h⁻¹ and 1%
20 gradient, the gradient was subsequently increased to 4% and speed increased by 1km·h⁻¹·min⁻¹
21 until the limit of volitional tolerance (P. I. Brown, Hughes, & Tong, 2008). Online breath by
22 breath gas analysis (MetaLyser II, Cortex Biophysik, Birmingham, UK) was used to
23 determine $\dot{V}O_{2peak}$, defined as the highest 30s $\dot{V}O_2$ recorded during the test.

1 During the second preliminary trial participants were familiarised with all testing
2 equipment and protocols. Participants were fitted with the 25 kg backpack (Web Tex,
3 Bedford, UK) which incorporated two shoulder straps and a waist strap which were adjusted
4 individually and recorded to the nearest mm for subsequent trials. Participants then
5 completed a full habituation of the experimental trial (see below). The habituation trial was
6 performed to minimise systematic error in the experimental time-trial tests as recommended
7 by our group previously (Faghy & Brown, 2014a). An absolute load mass was selected in
8 favour of a load relative to body mass to reflect training requirements of occupational groups
9 such as the Armed Forces and Emergency Services (Rayson, Holliman, & Belyavin, 2000).
10 During trials, the load mass was evenly distributed in the central compartment of the
11 backpack and worn in accordance with the manufactures guidelines.

12 *Experimental trial*

13 Although inherently difficult to devise a laboratory-based protocol which precisely reflects
14 the load, intensity, duration, terrain of occupational requirements the walking speed and
15 duration, rest period and the time-trial distance in this study were selected to reflect the
16 occupational demands of current British Army training and fitness assessments and was in
17 line with previous recommendations for laboratory-based studies of load carriage (Rayson et
18 al., 2000). Prior to a 6 week intervention (see below), participants completed a 2.4 km time
19 trial which was pre-loaded with 60 min walking exercise and a 15 min seated rest. Initially,
20 participants walked for 60 min, 0% gradient at $6.5\text{km}\cdot\text{h}^{-1}$ carrying a 25 kg backpack (hereon
21 referred to as the load carriage trial: LC). This phase of the experimental trial was consistent
22 with the British Army Infantry Basic Combat Fitness Test where soldiers carry a 25 kg load
23 at $15\text{ min}\cdot\text{mile}^{-1}$ pace ($6.4\text{ km}\cdot\text{h}^{-1}$) (The British Army, 2014). Following a 15 min seated
24 recovery which reflects the duration of a rest period on a military operation (e.g., while on a
25 patrol), participants completed a self-paced 2.4 km time-trial (hereon referred to as LC_{TT}).

1 The 2.4 km loaded time trial has been used previously to determine the physical and
2 physiological responses to acute changes in British Army infantry training programmes (P. E.
3 H. Brown et al., 2007, 2010) and is an assessment tool of the British Army Infantry
4 Advanced Combat Fitness Test (The British Army, 2014). Throughout LC_{TT} the speed of the
5 treadmill was manually adjusted by the participant in order to complete the distance in the
6 quickest time possible; the time elapsed was blinded from participants. In addition, we have
7 previously demonstrated (Faghy & Brown, 2014a) that this protocol provides an excellent
8 tool for assessing load carriage performance between days (log ratio limits of agreement
9 [LoA]; bias =1.02 [standard error 0.37], Confidence interval [CI] 0.18 to 1.86; random error
10 ratio =1.11 [standard error 0.65], CI lower LoA -0.49 to 2.43, CI upper LoA -0.33 to 2.59).
11 Following the intervention, participants performed two trials on separate days: 1) a shortened
12 re-familiarisation trial: 20 min LC at 6.5 km·h⁻¹, 0% gradient, and 15 min seated rest,
13 followed by 2.4 km LC_{TT} and 2) a post training experimental trial identical to the pre-
14 intervention experimental trial detailed above. The shortened re-familiarisation trial
15 performed has been shown to maximise reliability in between day load carriage assessments
16 (Faghy & Brown, 2014a).

17 *Equipment and Measurements*

18 Baseline pulmonary function and inspiratory ($P_{I_{max}}$) and expiratory ($P_{E_{max}}$) muscle
19 pressure were measured according to published guidelines (McConnell 2007). Briefly,
20 pulmonary function was assessed using an electronic flow sensor (MS03, Micro Medical,
21 Buckinghamshire, UK) calibrated according to the manufacturers guidelines with a 3-L
22 syringe. A hand-held mouth pressure meter (MicroRPM; CareFusion, Hampshire, UK)
23 measured $P_{I_{max}}$ and $P_{E_{max}}$, with manoeuvres initiated from residual volume and total lung
24 capacity, respectively and sustained for at least 1 s. During experimental trials, $P_{I_{max}}$ and
25 $P_{E_{max}}$ were measured at rest, immediately following LC and immediately following LC_{TT}.

1 Participants performed a minimum of 6 manoeuvres every 30 s and continued until 3 efforts
2 within ~10% of each other were observed, the highest value was used for analysis
3 (McConnell, 2007).

4
5 All non-respiratory variables were measured immediately prior to and during the final
6 minute of LC, immediately prior to, after 1.2km and during the final 0.1km of LC_{TT}. Heart
7 rate (HR) was measured using short-range telemetry (Polar T31, Kempele, Finland); Blood
8 lactate ($[\text{lac}^-]_{\text{B}}$; Accu-Check, Safe T-Pro, Birmingham, UK) and blood glucose concentrations
9 ($[\text{glucose}]_{\text{B}}$, Accutrend blood glucose, Birmingham, UK) were measured singularly, in whole
10 blood from 5 μ l arterialised-venous fingertip capillary samples. Expired pulmonary gases
11 were collected over a 60s period using a 2-way non re-breathing valve connected to a
12 Douglas bag via wide-bore tubing (Cranlea and Co, Birmingham, UK) and analysed using a
13 gas analyser (HITECH, GIR250, Luton, UK) and gas vacuum (Harvard, Apparatus, Cranlea
14 and Co, Birmingham, UK). Whole body perceived exertion (RPE) was measured using the
15 Borg scale where 6= no exertion and 20= maximal exertion. Scales of perception of effort
16 were used for leg (RPE_{legs}) and breathing ($\text{RPE}_{\text{breathing}}$) discomfort using a visual analogue
17 scale: where 0 = no exertion and 10 = maximal exertion (Verges et al., 2007).

18 *Training Intervention*

19 Participants were assigned to one of two training groups either a 1) pressure threshold
20 IMT or 2) a placebo control group (PLA), participants were assigned to groups via an
21 independent technician who conducted all calibration sessions (detailed below) to blind the
22 principal investigators to group allocation. Both groups were instructed by the independent
23 technician that they would receive an ergogenic effect from the training to maximise
24 motivation and expectation which was reinforced throughout the study. IMT was performed
25 using a commercially available pressure threshold inspiratory muscle training device

1 (POWERbreathe® classic series, HaB International, Warwickshire, UK). This device was
2 selected as it targets both axis of the force-velocity relationship, and unlike other available
3 methods pressure threshold training is flow-independent therefore sustaining the prescribed
4 resistance throughout each breath of the training regimen. The training load of the device
5 was individually calibrated at $\sim 50\%$ $P_{I_{max}}$ for the duration of the intervention and comprised
6 thirty consecutive dynamic inspiratory efforts, twice daily, for six weeks. Each inspiratory
7 effort was initiated from residual volume and participants endeavoured to maximise tidal
8 volume (P. I. Brown et al., 2014). To avoid hypocapnia participants were instructed after
9 each inspiratory effort to expire slowly and fully consequently reducing their breathing
10 frequency. Assessments of $P_{I_{max}}$ were completed at two week intervals during the training
11 intervention, allowing the training device to be adjusted to maintain training load. Training
12 adherence in both groups was monitored by completion of a training diary of which other
13 normal training activities throughout the intervention were also monitored. Regular
14 inspection of participant's training diaries confirmed that training remained unchanged
15 throughout the study. This specific training protocol is well documented to increase chest
16 wall inspiratory muscle and diaphragm strength (P. I. Brown et al., 2014; P. I. Brown,
17 Sharpe, & Johnson, 2010, 2012).

18 The PLA device was identical to the IMT device, however the resistance spring was
19 removed and replaced with loosely packed aquarium gravel with participants informed that
20 the gravel was oxygen absorbent thereby reducing $F_{I_{O_2}}$ to mimic altitude training.
21 Participants were instructed to breathe normally, while at rest, through the device for a period
22 of 15 min, once daily, five days wk^{-1} . As with the IMT device $P_{I_{max}}$ was assessed at two week
23 intervals during which time the PLA gravel was replaced. This PLA protocol has been well
24 documented to maximise participant expectation and motivation, yet cause no change in
25 inspiratory muscle strength or changes in time-trial performance (Johnson et al., 2007).

1 *Statistical Analysis*

2 Differences between groups for physical characteristics were assessed using an
3 independent t test. Between subject differences and interaction effects for dependent variables
4 were assessed using a 2 (group: IMT vs. PLA) x 3 (time point: rest, post-LC, post-LC_{TT}) x 2
5 (Trial pre vs. post intervention) mixed model ANOVA with Tukey's post hoc analysis using
6 SPSS for Windows (Chicago, IL, USA). A priori α was set at 0.05. All results are presented
7 as mean \pm SD. Effect size was calculated using Cohen's d ($d=(x^1-x^2)/\text{pooled } \sigma$), judgements
8 were made on the magnitude of the observed effect based on the 'minimal worthwhile effect'
9 as described in previous literature (Hopkins, 2000).

Results

1 *Group descriptive characteristics*

2 Pre-intervention baseline group descriptive characteristics of the participants are shown in
3 Table 1. There were no between group differences in any variable ($P>0.05$).

4 *Time-trial Performance*

5 Pre and post-intervention LC_{TT} time for both groups are shown in Tables 2 and 3. Time-
6 trial performance improved post-IMT (absolute reduction = 1.3 ± 0.7 min, $8 \pm 4\%$, group x
7 trial interaction effect: $p<0.05$; effect size: $d=0.93$), and was unchanged in PLA. The mean
8 speed during LC_{TT} increased after IMT (2.6 ± 0.3 m·s⁻¹ vs 2.8 ± 0.4 m·s⁻¹, $p<0.05$; effect size:
9 $d=0.86$) and was unchanged after PLA (pre 2.5 ± 0.3 m·s⁻¹ vs post 2.5 ± 0.4 m·s⁻¹; $p>0.05$).

10 *Maximal Inspiratory and Expiratory Pressures*

11 Training compliance was high in both groups (IMT $94 \pm 8\%$ and PLA $91 \pm 11\%$) and was
12 similar to previous IMT studies using the same protocol (P. I. Brown et al., 2012; Johnson et
13 al., 2007). Resting values and changes between pre and post-intervention for $P_{I_{max}}$ and $P_{E_{max}}$
14 are shown in Figures 1 and 2, respectively. Prior to the intervention, relative to resting values,
15 $P_{I_{max}}$ and $P_{E_{max}}$ were reduced post-LC and post LC_{TT} in both groups (pooled data, $P_{I_{max}}$: $13 \pm$
16 7% and $16 \pm 8\%$. $P_{E_{max}}$: $14 \pm 7\%$ and $17 \pm 16\%$; $p<0.05$) which was similar post intervention
17 in PLA. Relative to pre-IMT, $P_{I_{max}}$ was greater at each time-point: rest ($+31\%$, $p<0.05$), post-
18 LC ($+19\%$, $p<0.05$) and post-LC_{TT} ($+18\%$, $p<0.05$). Relative to pre-IMT, $P_{E_{max}}$ was also
19 greater at each time-point: rest ($+17\%$, $p<0.05$), post-LC ($+15\%$, $p<0.05$) and post-LC_{TT}
20 ($+7\%$, $p<0.05$). However, although $P_{I_{max}}$ was greater at each time point after IMT, the
21 reductions relative to resting values were similar to pre-intervention after the 60 min steady
22 state phase (pre IMT $\% \Delta P_{I_{max}}$: $11.3 \pm 6.8\%$ vs post IMT $12.8 \pm 10.8\%$; $p>0.05$) and the time
23 trial (pre IMT $\% \Delta P_{I_{max}}$: $2.3 \pm 10.2\%$ vs post IMT $5.3 \pm 12.7\%$; $p>0.05$). Reductions in $P_{E_{max}}$
24 were greater compared with pre-intervention (pre IMT $\% \Delta P_{E_{max}}$: $5.5 \pm 7.8\%$ vs post IMT

1 11.7 ± 7.8%; $p < 0.05$) and similar post LC_{TT} (pre IMT %Δ $P_{E_{max}}$: 18.6 ± 6.2 % vs post IMT
2 21.6 ± 12.2%; $p > 0.05$).

3

4 Pulmonary Function

5 Baseline measures of pulmonary function are shown in Tables 2 and 3. Baseline and
6 changes between trial were similar between groups ($p < 0.05$) prior to and post intervention
7 and remained unchanged in both groups, following the intervention.

8 *Pulmonary Gas Exchange*

9 Gas exchange responses to the experimental trial for the IMT and PLA groups are shown
10 in Tables 2 and 3, respectively. Prior to the intervention with the exception of RER and one
11 measure of $\dot{V}O_2$ (post-LC_{TT}, pre-intervention, IMT group) minute ventilation (\dot{V}_E), $\dot{V}O_2$ and
12 carbon dioxide production ($\dot{V}CO_2$) was greater than rest post-LC and post-LC_{TT} ($p < 0.05$),
13 with no between group differences ($p > 0.05$). Following the intervention, similar values were
14 observed at baseline and over time for all variables in both groups.

15

16 *Physiological Responses*

17 Baseline values of HR were similar between groups at rest (Table 2 and 3). HR increased
18 from rest, post-LC (pooled data: post-LC, 31 ± 10%; $p < 0.01$) and post-LC_{TT} (pooled data: 45
19 ± 8%; $p < 0.01$) and also increased between post-LC to post-LC_{TT} (23 ± 9%; $p < 0.05$); prior to
20 the intervention HR was not different between groups ($p > 0.05$). Following the intervention
21 HR was significantly lower (7%) post-LC in IMT ($p < 0.05$), relative to the same time point
22 and lower (6%) than PLA ($p < 0.05$). Both $[lac^-]_B$ and $[Glucose]_B$ were similar between groups
23 at rest and over time before and after the intervention ($p > 0.05$).

1 *Perceptual Responses*

2 Changes in perceptual responses for IMT and PLA are shown in Tables 2 and 3,
3 respectively. Prior to the intervention all perceptual responses were similar at baseline
4 between groups ($p>0.05$), and increased from rest, both at post-LC ($p<0.01$) and post-LC_{TT}
5 ($p<0.01$) with no differences between groups ($p>0.05$). Post intervention responses were
6 similar in PLA but whole-body RPE (8%) and RPE_{legs} (10%) were lower post-LC in IMT
7 ($p<0.05$).

Discussion

1 The aim here was to investigate the effects of 6-wks IMT on pre-loaded time-trial
2 performance while carrying a 25 kg thoracic load in a backpack. The novel findings were
3 threefold 1) LC_{TT} performance was improved, 2) despite an increase in respiratory muscle
4 strength post IMT, respiratory muscle fatigue was unchanged relative to pre-intervention
5 values and 3) heart rate and perceived exertion during exercise were attenuated during LC
6 after IMT. Accordingly, this study is the first to demonstrate an ergogenic effect of IMT
7 using a double-blind placebo controlled design during self-paced exercise while carrying a
8 thoracic load.

9 *Time-trial Performance*

10 Pre-intervention LC_{TT} performance in both groups was consistent with previous literature
11 using the same protocol (Faghy & Brown, 2014a, 2014b). In this double-blind placebo
12 controlled study (see *intervention* above for more details); time-trial performance was
13 significantly improved following IMT by $8 \pm 4\%$ with no change in PLA. When combined
14 with the significant interaction effect and a very large effect size ($d=0.93$); these results are
15 regarded as a worthwhile effect (Hopkins, 2000). To our knowledge there is no other data
16 available to compare this IMT-mediated improvement in load carriage performance.
17 Accordingly, our findings must be compared with data from un-loaded exercise (i.e., no
18 backpack). IMT has repeatedly improved whole-body performance in cycling, running,
19 rowing and repeated sprint exercise with improvements of 3 to 6 % (for a review see
20 HajGhanbari et al., 2013; Ili et al., 2012). Given the improvement we observed here we have
21 demonstrated for the first time that IMT has a genuine ergogenic effect upon 25 kg load
22 carriage exercise performance.

1 *Respiratory Muscle Pressure*

2 The mechanism(s) accounting for the improvement in performance are not well
3 understood but are likely explained by improved inspiratory muscle strength and its effect
4 upon cardiorespiratory and/or perception of effort interactions during exercise. Inspiratory
5 muscle strength increased post-IMT ($31 \pm 26\%$), similar to previous work (Johnson et al.,
6 2007). Prior to the intervention we observed significant inspiratory muscle fatigue post-LC
7 and LC_{TT} which is consistent with previous studies using the same protocol (Faghy & Brown,
8 2014a, 2014b) and with firefighters exercising with SCBA (Butcher et al., 2006). Post-IMT,
9 $P_{I_{max}}$ was greater at each time point throughout the experimental trial; however, the
10 magnitude of inspiratory muscle fatigue was similar. This is in contrast to previous studies
11 that show attenuated inspiratory muscle fatigue after cycling time trial exercise (Romer,
12 McConnell, & Jones, 2002a). Our findings are however in agreement with Verges et al
13 (2006), who demonstrate similar rates of respiratory muscle fatigue following endurance-
14 focused respiratory muscle training despite a 272% improvement in respiratory muscle
15 endurance. This was attributed to either 1) an ineffective intervention, this is not the case here
16 as we observed large changes in $P_{I_{max}}$ with no change in the PLA group or 2) that IMT failed
17 to attenuate any inspiratory muscle fatigue associated with non-respiratory roles such as
18 supporting posture and spinal stability. The latter, may explain our findings as wearing a
19 backpack increases postural sway and the reliance upon the diaphragm to stabilise the spine
20 (Hodges, Butler, McKenzie, & Gandevia, 1997). In addition, load carriage fatigues accessory
21 respiratory muscles of the thorax which support pulmonary ventilation during exercise with
22 high breathing demands (Blacker, Fallowfield, Bilzon, & Willems, 2010). Therefore it is
23 possible that IMT failed to attenuate fatigue that is associated with the non-respiratory
24 functions of the inspiratory muscles, although future research is required to address this.
25 Despite, no reduction in respiratory muscle fatigue after IMT, we observed significant IMT-

1 mediated changes in cardiovascular strain and the perceptions of effort which we attribute to
2 the increase in inspiratory muscle strength.

3 *Cardiovascular Strain*

4 We observed a 7% reduction in HR post-LC after IMT, indicating reduced cardiovascular
5 strain. Reduced cardiovascular strain during loaded breathing tasks has been demonstrated
6 previously after IMT (lowered HR and mean arterial pressure) attributed to increases in
7 strength. This facilitates oxidative adaptations within the inspiratory muscles (Witt, Guenette,
8 Rupert, McKenzie, & Sheel, 2007) permitting them to contract at a lower relative intensity
9 (Turner et al., 2012). Here, this was attributed to an attenuated sympathetic efferent response
10 (i.e. reduced metaboreflex activation) to fatiguing inspiratory muscle work whilst at rest.
11 Consequently, after IMT in the present study, reduced cardiovascular strain due to a similar
12 mechanism prior to LC_{TT} may have contributed to the improved time trial performance. The
13 reduction in cardiovascular strain observed in this study may also be explained in part, by
14 normalised intrathoracic pressures swings during the breathing cycle (Miller, Beck, Joyner,
15 Brice, & Johnson, 2002). Load carriage increases gastric pressure which in turn elevates
16 central venous and transdiaphragmatic pressure which in turn alters cardiac pre- and after-
17 load, affecting both cardiac output and heart rate (Miller et al., 2002). Following IMT we
18 observed an increased P_{Emax} which may also permit the expiratory muscles to operate at a
19 lower relative intensity during exercise facilitating venous return (Butcher et al., 2006) and
20 normalising this cardiovascular response.

21

1 *Perceptual Responses*

2 We reported a ~4% reduction in breathing discomfort and a ~10% reduction in leg
3 discomfort during LC following IMT; similar to previous observations during rowing and
4 cycling time trial exercise (Romer et al. 2002a; Volianitis et al. 2001) and isolated breathing
5 challenges (Verges et al, 2007). In the present study, these reductions were independent of
6 exercise intensity and ventilatory demand since LC intensity was fixed during LC and
7 produced a similar ventilatory response (Tables 2 and 3). IMT has been shown to reduce the
8 oxygen cost of breathing for a given fixed pulmonary ventilation (Turner et al., 2012).
9 Therefore, repeated bouts of IMT may alter the discharge frequency of mechano-sensitive
10 type III and IV nerve afferents due to lower metabolic demand, reducing afferent feedback
11 and the perception of breathing discomfort (Sinoway et al. 1996). It is possible that the
12 reduction in perception of effort is linked to the attenuation of the metaboreflex whereby limb
13 blood flow is sustained and limb fatigue reduced since IMT increases the threshold for
14 activation of this response (McConnell & Lomax, 2006). Recent evidence suggests that limb
15 discomfort induced by fatiguing knee extensor exercise heightens the perception of breathing
16 discomfort with no change in pulmonary ventilation (Grippo et al., 2010; Sharma, Morris, &
17 Adams, 2015). Therefore, improved locomotor muscle function following IMT due to
18 improved limb blood flow (attenuated metaboreflex) would attenuate limb discomfort and
19 also subsequently breathing discomfort. Despite this notion, during sub-maximal exercise
20 (LC: 59% $\dot{V}O_{2peak}$) any reductions in limb blood flow are likely to be compensated by
21 increased locomotor muscle oxygen extraction (Jones et al., 2011; Romer, Haverkamp,
22 Lovering, Pegelow, & Dempsey, 2006). IMT-mediated reductions in breathing discomfort are
23 correlated with improved exercise tolerance (Romer, McConnell, & Jones, 2002b) therefore
24 regardless of the mechanism(s) afferent discharge, whether originating within the locomotor
25 and/or respiratory musculature are incredibly important, as they both project to the

1 sensorimotor cortex which regulates central motor drive (Amann & Dempsey, 2008).
2 Accordingly a reduction in afferent feedback from these sources would improve central
3 motor drive for the subsequent time trial effort.

4 *Methodological limitations*

5 We employed volitional measures of respiratory muscle force here, which do not directly
6 reflect the force of the diaphragm; rather, P_{Imax} reflects the volitional force of all inspiratory
7 muscles working in synergy and may not be sufficiently sensitive to rule out respiratory
8 fatigue changes. P_{Imax} and trans-diaphragmatic pressure are correlated before and after IMT
9 (P. I. Brown et al., 2014) therefore we suggest that our measures of ΔP_{Imax} provide a useful
10 measure of inspiratory muscle force and a useful surrogate of diaphragm function. In
11 addition, although non-volitional measures of muscle force are preferred by measuring the
12 oesophageal, gastric and hence transdiaphragmatic pressure response to electrical or magnetic
13 stimulation, due to technical limitations volitional measures were employed. Consequently, in
14 line with our previous work (Faghy & Brown, 2014a) we spent time ensuring familiarisation
15 with this measurement and strove to maximise motivation throughout. It is also important to
16 note that volitional measures have superior between day (i.e., pre to post intervention)
17 reproducibility (Romer & McConnell, 2004) which in combination with their use in previous
18 literature justifies their use in this study.

19 *Conclusion*

20 Inspiratory muscle training improved 2.4 km time trial performance when exercise was
21 performed with 25 kg thoracic load carriage and pre-loaded with 60 min sub-maximal
22 exercise. These findings are most likely explained by an increase in inspiratory and
23 expiratory muscle strength and the subsequent effects upon cardiovascular strain and the

- 1 sensations of the perception of effort. These findings may have implications for both
- 2 occupational and recreational groups where load carriage is a critical role-related task.

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2

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4

5 **Disclosure Statement**

6

7 Ethics Standards

8 All experimental procedures and methods of assessment used in this study were approved by

9 the host universities ethics committee and conform to the laws of the United Kingdom.

10

11 Conflicts of interest:

12 No conflicts of interest for each of the authors.

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Tables

Table 1. Mean \pm SD for the descriptive characteristics of the participants (n=19).

Descriptive Characteristic	IMT (n=10)	PLA (n=9)
Age (years)	25.8 \pm 8.7	24.2 \pm 6.3
Body Mass (kg)	77.4 \pm 9.3	78.1 \pm 7.7
Height (m)	1.74 \pm 0.04	1.81 \pm 0.08
Body Fat (%)	21.9 \pm 6.1	24.4 \pm 5.1
Lean body mass (kg)	59.6 \pm 5.6	58.0 \pm 4.7
Fat mass (kg)	17.8 \pm 6.0	20.1 \pm 5.3
Bone mineral density (g/cm ²)	1.3 \pm 0.2	1.3 \pm 0.2
Bone mineral content (kg)	3.2 \pm 0.5	3.2 \pm 0.6
$\dot{V}O_{2peak}$ (L·min ⁻¹)	4.3 \pm 0.5	4.1 \pm 0.4
$\dot{V}O_{2peak}$ (ml·kg ⁻¹ ·min ⁻¹)	52.6 \pm 5.7	49.2 \pm 4.5
Training Adherence (%)	93.8 \pm 7.9	90.5 \pm 11.2

$\dot{V}O_{2peak}$; maximal oxygen uptake in absolute units and relative to body mass.

Table 2. Responses to 60 min load carriage (LC) and 2.4 km load carriage time trial (LC_{TT}) prior to and following inspiratory muscle training (IMT) (n=10).

Variable	Pre-IMT			Post-IMT		
	Pre-LC	Post-LC	Post-TT	Pre-LC	Post-LC	Post-TT
Time-trial (min)	-	-	15.45 ± 1.70	-	-	14.39 ± 1.80 ^{C*}
FEV ₁ (L)	3.7 ± 0.4	3.6 ± 0.5	3.7 ± 0.4	3.7 ± 0.5	3.6 ± 0.4	3.6 ± 0.5
FVC (L)	4.7 ± 0.5	4.6 ± 0.6	4.6 ± 0.5	4.7 ± 0.5	4.5 ± 0.6	4.4 ± 0.8
FEV ₁ / FVC (%)	79 ± 8	78 ± 10	81 ± 11	79.2 ± 10	80.6 ± 11	83 ± 10
PEF (L·min ⁻¹)	492 ± 105	474 ± 106	517 ± 86	525.1 ± 111	498.3 ± 71	503 ± 82
HR (beats·min ⁻¹)	102 ± 16	145 ± 19 ^A	187 ± 15 ^{AB}	108 ± 15	135 ± 29 ^{A*C}	193 ± 7 ^{AB}
[Lac ⁻] _B (mmol·l ⁻¹)	1.5 ± 1.1	1.3 ± 0.6	8.5 ± 4.2 ^{AB}	1.5 ± 0.4	1.3 ± 0.6	9.6 ± 3.2 ^{AB}
\dot{V}_E (L·min ⁻¹)	40.8 ± 8.2	46.2 ± 9.2 ^A	62.4 ± 22.6 ^{AB}	37.2 ± 11.3	46.6 ± 6.7 ^A	79.1 ± 25.5 ^{AB}
$\dot{V}O_2$ (L·min ⁻¹)	1.49 ± 0.35	1.66 ± 0.30	1.17 ± 0.54	1.27 ± 0.40	1.74 ± 0.31 ^A	1.68 ± 0.76
$\dot{V}CO_2$ (L·min ⁻¹)	1.40 ± 0.28	1.72 ± 0.27 ^A	1.71 ± 0.55	1.14 ± 0.43	1.60 ± 0.29 ^{AB}	2.06 ± 0.51 ^{AB}
RER	0.95 ± 0.14	1.05 ± 0.15	1.64 ± 0.75	0.89 ± 0.21	0.93 ± 0.12	1.33 ± 0.33 ^A
RPE (AU)	7 ± 1	12 ± 2 ^A	16 ± 3 ^{AB}	6 ± 0	11 ± 3 ^{A*}	17 ± 3 ^{AB}
RPE _{legs} (AU)	0 ± 0	4 ± 1 ^A	7 ± 1 ^{AB}	0 ± 0	3 ± 2 [*]	7 ± 2 ^{AB}
RPE _{breathing} (AU)	0 ± 0	3 ± 2 ^A	7 ± 2 ^{AB}	0 ± 0	3 ± 2 ^A	8 ± 2 ^{AB}

Maximum inspiratory pressure ($P_{I_{max}}$), maximum expiratory pressure ($P_{E_{max}}$), forced vital capacity (FVC), forced expiratory volume in one second (FEV₁), forced expiratory volume in one second / forced vital capacity ratio (FEV₁/ FVC), peak expiratory flow (PEF): Heart rate (HR), Blood Lactate [Lac⁻]_B, minute ventilation (\dot{V}_E), oxygen consumption ($\dot{V}O_2$), carbon dioxide production ($\dot{V}CO_2$), Respiratory exchange ratio (RER), Arbitrary units (AU). ^A = different to baseline $p < 0.05$, ^B = different to post-LC $p < 0.05$, ^C = different between groups ^{*} = different to pre.

Table 3. Responses to 60 min load carriage (LC) and 2.4 km load carriage time trial (LC_{TT}) prior to and following PLA (PLA, n=9).

Variable	Pre-PLA			Post-PLA		
	Pre-LC	Post-LC	Post-TT	Pre-LC	Post-LC	Post-TT
Time-trial (min)	-	-	16.44 ± 2.06	-	-	16.65 ± 2.53
FEV ₁ (L)	4.3 ± 0.5	3.9 ± 1.5 ^A	4.2 ± 0.6	4.5 ± 0.4	4.2 ± 0.6	3.9 ± 0.7
FVC (L)	5.2 ± 0.6	5.0 ± 0.9	4.8 ± 0.9	5.5 ± 0.6	5.0 ± 0.7	4.4 ± 0.9
FEV ₁ / FVC (%)	84 ± 8	85 ± 11	87 ± 11	81 ± 9	85 ± 12	88 ± 9
PEF (L·min ⁻¹)	559.1 ± 76	539 ± 90	537 ± 79	570 ± 60	557 ± 95	523 ± 95
HR (beats·min ⁻¹)	96 ± 10	140 ± 20 ^A	190 ± 11 ^{AB}	103 ± 13	144 ± 20 ^A	184 ± 23 ^{AB}
[Lac ⁻] _B (mmol·l ⁻¹)	1.7 ± 0.5	1.8 ± 1.1	8.8 ± 2.8 ^A	1.7 ± 0.5	1.6 ± 0.5	8.5 ± 3.1 ^A
\dot{V}_E (L·min ⁻¹)	39.5 ± 7.5	50.6 ± 6.5 ^A	61.0 ± 23.9 ^{AB}	34.2 ± 12.2	48.6 ± 5.3 ^A	78.1 ± 30.1 ^{AB}
$\dot{V}O_2$ (L·min ⁻¹)	1.65 ± 0.30	1.82 ± 0.15	1.44 ± 0.79	1.31 ± 0.57	1.76 ± 0.23 ^A	1.52 ± 0.28
$\dot{V}CO_2$ (L·min ⁻¹)	1.46 ± 0.26	1.76 ± 0.26	1.74 ± 0.77	1.14 ± 0.48	1.64 ± 0.30 ^A	2.03 ± 0.70 ^{AB}
RER	0.89 ± 0.11	0.97 ± 0.15	1.30 ± 0.38 ^A	0.93 ± 0.21	0.95 ± 0.10	1.32 ± 0.35 ^{AB}
RPE (AU)	6 ± 1	11 ± 3 ^A	17 ± 2 ^{AB}	7 ± 1	11 ± 3 ^A	17 ± 3 ^{AB}
RPE _{legs} (AU)	0 ± 0	3 ± 2	7 ± 2 ^{AB}	0 ± 1	3 ± 2	8 ± 2 ^{AB}
RPE _{breathing} (AU)	0 ± 0	2 ± 2	7 ± 1 ^{AB}	0 ± 1	2 ± 2	8 ± 2 ^{AB}

Maximum inspiratory pressure ($P_{I_{max}}$), maximum expiratory pressure ($P_{E_{max}}$), forced vital capacity (FVC), forced expiratory volume in one second (FEV₁), forced expiratory volume in one second / forced vital capacity ratio (FEV₁/ FVC), peak expiratory flow (PEF): Heart rate (HR), Blood Lactate [Lac⁻]_B, minute ventilation (\dot{V}_E), oxygen consumption ($\dot{V}O_2$), carbon dioxide production ($\dot{V}CO_2$), Respiratory exchange ratio (RER), Arbitrary units (AU).^A = different to baseline $p < 0.05$, ^B = different to post-LC $p < 0.05$, .

Figure Captions

Figure 1. Between and within group changes in $P_{I_{max}}$ (panel A) and $P_{E_{max}}$ (panel B) prior to and following inspiratory muscle training. The diamond markers represent the IMT training group and the squares represent the placebo groups, values are presented as mean \pm sd. ^A, different to baseline; ^B, different to Post-LC; *, Different between groups.