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SPORTS PERFORMANCE

Mental fatigue independent of boredom and sleepiness does not impact self-paced physical or cognitive performance in normoxia or hypoxia

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ABSTRACT

This study investigated the individual and combined effects of mental fatigue (MF) and hypoxia (HYP) on physical and cognitive performance. Fifteen males (24 ± 3 years) completed one familiarization session and six experimental trials, including: 1) normoxia (0.209 FiO_2) and no MF; 2) normoxia (0.209 FiO_2) with MF; 3) mild normobaric HYP (0.13 FiO_2) and no MF; 4) mild normobaric HYP (0.13 FiO_2) with MF; 5) severe normobaric HYP (0.10 FiO_2) and no MF; 6) severe normobaric HYP (0.10 FiO_2) with MF. Each condition included a 15-min self-paced time trial, followed by a 60-s isometric maximal voluntary contraction of the biceps brachii. MF was induced using a 16-min individualized cognitive test prior to exercise performance. Following each time trial, participants performed the Tower of Hanoi cognitive test. A main effect of HYP was observed on average power output, oxygen consumption and muscle oxygenation ($P \leq 0.004$), with no effect of MF ($P \geq 0.599$). Voluntary activation of the biceps brachii was also reduced in HYP (68.42 ± 5.64%, $P = 0.039$). No effect of MF or HYP was observed on cognitive performance ($P \geq 0.138$). HYP impacted physical performance, whilst MF had no effect on self-paced physical or cognitive performance.

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Mental fatigue; hypoxia; combined stressors; cognitive performance; physical performance

Introduction

At altitude, the atmospheric partial pressure of oxygen (PO_2) reduces in proportion to the barometric pressure, which results in a progressive reduction in the partial pressure of oxygen in alveolar air, arterial blood and ultimately muscle tissue (Brown & Grocott, 2013). The result is an inability to sustain prolonged exercise performance at high-altitude (Fulco et al., 1998; Wehrlin & Hallén, 2006). While cardiac output increases to partially compensate decreases in arterial oxygen saturation (Fulco et al., 1998; Roth et al., 2002), the lower driving pressure for oxygen at the muscle tissue, as well as the shift to greater percentages of maximum oxygen uptake (VO_{2max}), necessitates an increase in muscle fibre recruitment to sustain a given workload (Amann and Calbet 2008). Combined with lower oxygen availability in the brain, increases in both central and peripheral fatigue have been observed (Goodall et al., 2012). Moderate altitude has been shown to decrease dynamic muscle endurance as a result of peripheral fatigue interfering with the muscles' force and power generating capacity (Lloyd et al., 2015, 2016), while at high-altitude, central fatigue induced by suboptimal performance of the motor cortex is suggested to be the primary factor impacting performance (Goodall et al., 2012). Indeed, a negative impact of hypoxia on the processing abilities of the brain have been observed, including performance reductions in, e.g., memory recall, attention, verbal ability, perception, learning and spatial ability, and cognitive flexibility (Abraini et al., 1998; Griva et al., 2017; Kramer et al., 1993; McMorris et al., 2017; Taylor et al., 2016), the extent of which is largely dependent on the level of arterial partial pressure of oxygen with lower levels (<60 mmHg) predicting reduced cognitive performance, independent of whether the hypoxia is normobaric (i.e., simulated) or hypobaric (i.e., altitude) (McMorris et al., 2017).

Mental fatigue, a result of overloading the working memory capacity (Borragán et al., 2017) and characteristic of high-altitude occupations (Kottke et al., 2015), is a common cause of accidents and errors (Tanaka, 2015), as well as being detrimental to performance and hence survival in environmental extremes. Briefly, mental fatigue has been defined as a psychobiological state, characterized by decreased cognitive performance, focus, motivation and mood disturbances (Marcora et al., 2009; McMorris et al., 2018; Van Cutsem., 2017a). Critically, mental fatigue is a psychological state that is independent of other psychological constructs, such as sleepiness and boredom (O'Keeffe et al., 2019; Trejo et al., 2015). Negative implications of mental fatigue have been demonstrated on both physical and cognitive performance (Holtzer et al., 2011; Marcora et al., 2009; Smith et al., 2016; Van Cutsem et al., 2017a). However, not all research has observed an effect on physical and cognitive performance, with factors such as the perception of effort, motivation, cognitive fatigue protocol, and primary modulating mechanisms of performance (e.g., pacing strategies, peripheral fatigue), as suggested reasons for this discrepancy (Martin et al., 2015; Pageaux et al., 2013; Vrijkotte et al., 2018). In addition, research has also questioned the existence and significance of mental fatigue due to the small effect sizes observed and lack of clarity surrounding its impact (McMorris et al., 2018).

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Working in extreme environments is characterized by a combination of environmental and psychological stressors. At altitude, humans are exposed to environmental stressors such as hypobaric hypoxia, cold, and solar radiation, often in combination with, e.g., prolonged cognitive effort, anxiety and sleep deprivation (Bhaumik et al., 2008; Lloyd & Havenith, 2016). Despite the extensive presence of multifactorial environments, limited research has attempted to understand the underpinnings of combined physiological and psychological stressors on human physical and cognitive capacity (Tipton, 2012). The present investigation compared three levels of hypoxia (altitude) in combination with, and independently of mental fatigue, on both physical and cognitive performance. Three hypotheses were constructed: 1) As the environmental oxygen concentration reduces, both physical and cognitive performance will decline; 2) mental fatigue, independently of hypoxia, will negatively impact both physical and cognitive performance; and 3) as the level of hypoxia increases the impact of mental fatigue will also be reduced. Hypothesis 3 was constructed on the basis of a proposed combined stressors theory by Lloyd and Havenith (2016) which states that when stressors are combined, individual stressors may take precedence and reduce the impact of the other. Therefore, in the current research, it was hypothesized that during combined exposure to two stressors, i.e., mental fatigue and hypoxia, hypoxia would take precedence and reduce the impact of mental fatigue on physical and cognitive performance as the severity of hypoxia increases.

Methods

Participants and general procedures

Participants comprised of volunteers from Loughborough University. Fifteen physically active males aged 24 ± 3 years, with no history of neurological injury were recruited. Participants were paid for their participation pro-rata (£10 for the familiarization session, and £15 per main experimental trial) The study was approved by the Loughborough University ethics committee (R17-P129) and conformed to the standards set by the Declaration of Helsinki (World Medical Association, 2013), except for registration in a database. All experiments were conducted at the Environmental Ergonomics Research Centre, Loughborough, UK. Participants were requested not to consume any food or caffeine for up to 2-hours prior to participation in the study. Trials were counterbalanced using the Latin Square procedure where the six experimental conditions were considered independent. Participants were required to come in at the same time of day for each trial. Before participating in any procedures, participants provided written informed consent and were given the opportunity to discuss the study with the lead researcher. In total, the present study consisted of seven trials, in which six main experimental trials followed an initial familiarization session.

Familiarization session

The familiarization session constituted a critical part of the experimental procedures in this study. This is because a 16-min individualized Time load Dual-back (TloadDback) (Borragán et al., 2017) cognitive test was used to induce mental fatigue. The familiarization session was used to practice, and subsequently individualize the cognitive test, such that task difficulty matched each participant's own cognitive ability. The training and individualization process for the TloadDback, and its relationship to mental fatigue relative to other cognitive tests, has been described in detail by O'Keeffe et al. (2019) as well as by Borragán et al. (2017). A full overview of the TloadDback test is also provided in the section: "Mental Fatigue Inducement".

Following the individualization process for the TloadDback, participants were accustomed to the experimental procedures and a full rehearsal of the physical performance test was completed. The physical performance test included a self-paced 15-min time trial, as well as a series of brief and sustained isometric neuromuscular tests, including supramaximal nerve stimulation. Full details of the physical performance testing are provided at section: "Physical Performance Protocol". To complete the session, participants practised the cognitive performance test the "Tower of Hanoi" (TOH; disk number = 4; total moves = 15). The Tower of Hanoi is a measure of mental flexibility and working memory and a detailed description can be found at Humes et al. (1997). At the familiarization session, this cognitive test was repeated by participants until three successful (0% errors) consecutive performances were achieved.

Experimental overview

The present study consisted of six (FiO_2 x 3; MF x 2) main experimental conditions. The main experimental trials differed only by environmental oxygen concentration (0.209 vs 0.13 vs 0.10 FiO_2) and the presence of mental fatigue inducement (with vs without pre-test mental fatigue). The conditions were: NC: normoxia and no mental fatigue; NF: normoxia with mental fatigue; MC: mild normobaric hypoxia (0.13 FiO_2) and no mental fatigue; MF: mild normobaric hypoxia (0.13 FiO_2) with mental fatigue; SC: severe normobaric hypoxia (0.10 FiO_2) and no mental fatigue; SF: severe normobaric hypoxia (0.10 FiO_2) with mental fatigue. Normobaric hypoxia was manipulated using the inbuilt hypoxic air generator in the environmental chambers (T.I.S. S Peak Performance, Series 2009 Climate Chambers). A Servomex gas analyser (570A, Sussex, UK) was used to monitor the oxygen level inside the chamber, as well as the inbuilt analyser in the chamber. All conditions were completed at 21°C with 50% relative humidity.

In each of the main experimental trials, participants completed a protocol that consisted of: i) a pre-test re-familiarization of the Tower of Hanoi cognitive test; ii) a pre-test re-familiarization of the neuromuscular testing, including

supramaximal nerve stimulation; iii) completion of a pre-test mental fatigue inducement or control condition set-up as detailed by O'Keeffe et al. (2019); iv) a physical performance test, including a 15-min, self–paced, arm-bike performance test, immediately followed by a 60-sec isometric maximal voluntary contraction (MVC) of the biceps brachii with supramaximal nerve stimulation; and lastly, v) a timed attempt at the TOH cognitive test. A schematic of the main experimental trials are presented in Figure 1.

Mental fatigue inducement

A detailed description of using the TloadDback cognitive test to induce mental fatigue is described by O'Keeffe et al. (2019). The primary aim of the task is to be cognitively demanding and induce mental fatigue in a short period of time, whilst maintaining arousal. Briefly, the TloadDback test is a dual-task test, lasting 16-min, in which numbers and letters are alternated in sequence. The TloadDback, therefore, includes: a primary task (parity judgement test), where participants must distinguish between odd and even numbers; and a concurrent secondary task (classic n-back paradigm), in which a series of letters are presented, and participants respond when the letter presented is identical to the letter that was presented immediately before. As noted above, the TloadDback is also individualized, such that the speed at which the numbers and letters are presented at are standardized to an individual's cognitive ability, inducing a high cognitive workload which is required for inducing mental fatigue effectively using this test (Borragán et al., 2017). The individual stimulus detection time (i.e., shortest stimulus speed a person can process the incoming information at) was fixed at a speed that participants were no longer able to maintain a dual-task accuracy of 85% (Borragán et al., 2017). Motivation was manipulated through offering a monetary incentive, whereby participants were falsely informed that they would receive an additional £25 if their performance was in the top 50% of participants in the cognitive test. This deception protocol was incorporated to sustain motivation throughout performance of the TloadDback test in order to ensure the maintenance of cognitive effort, by limiting the impact of reduced self-control (Inzlicht et al., 2014). All pre-performance TloadDback tests were conducted on a standard desktop computer (Hewlett-Packard). The height of the monitor display (LA2205wg, 22 in., 1680 × 1050 resolution) was adjusted for each participant so that the stimulus appeared at eye level. All test stimuli (letters and numbers) appeared in white font against a black background.

Detailed physical performance protocol

All exercise testing was conducted on a custom-made arm-bike rig, built by the Environmental Ergonomics Research Centre. The device consisted of a seat and chest harness, a hand bike ergometer (Lode, NL) located directly in front of the participant, and a right arm support with an adjustable force transducer and wrist strap. The device was developed for measuring dynamic arm-bike performance, as well as contractile performance of the biceps brachii. This mode of exercise was chosen as the use of the upper body extremities (i.e., steep scrambling, technical climbing, pushing and pulling loads) and self-regulated exercise (i.e., time trial performance) is characteristic of work in altitude. Further, the ease and speed of transition to the custom-made MVC rig (i.e., <30-sec) after finishing the 15-min time trial was important to ensure rapid, optimal and consistent neuromuscular assessment.

Upon entering the chamber, participants practised a series of 3-sec MVC's with supramaximal nerve stimulation (see below). Following this, participants then conducted a 15-min, self–paced, arm-bike physical performance test in each of the test conditions. Participants were instructed to produce as much work as possible (i.e., Watts). No motivation was given but participants were able to see their power output and were informed when they had completed 5-min, 10-min and had

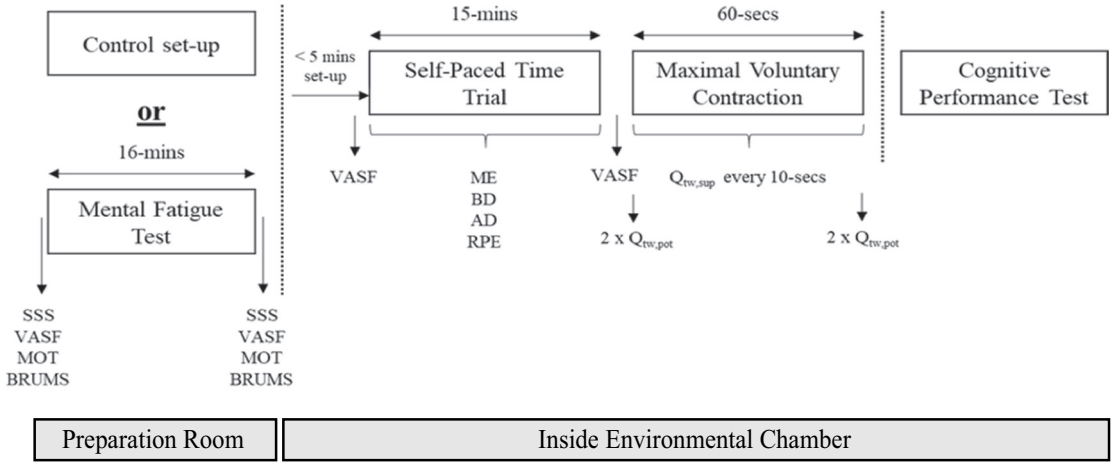


Figure 1. Schematic of main trials. VASF (Visual Analogue Scale of Fatigue), SSS (Stanford Sleepiness Scale), MOT (Motivation Scale), BRUMS (Brunel Mood Scale), ME (Mental Effort, Borg CR-100), BD (Breathing Discomfort, Borg CR-10), AD (Arm Discomfort, Borg CR-10), RPE (Rating of Perceived Exertion, Borg 6–20), SaO_2 (Oxygen Saturation), MVC (Maximal Voluntary Contraction), $Q_{tw,pot}$ (resting potentiated twitch), $Q_{tw,sup}$ (superimposed twitch). The set-up control and mental fatigue test were conducted in the preparation rooms outside the environmental chamber, whilst the physical and cognitive performance protocols were conducted inside the environmental chamber.

2-min remaining. The resistance factor on the Lode Angio Arm Bike was calculated based on the linear relationship with a traditional monarch brake weight, which is defined as; resistance factor alpha = 0.0161*brake weight + 0.0008. The brake weight used for this experiment was based on pre-experimental piloting and corresponded to 1% of participant's familiarization body mass (74.67 ± 9.74 kg). This was immediately followed by a 60-sec MVC with simultaneous nerve stimulation (see below), every 10-sec. Participants were harnessed around the shoulders and waist for the duration of the experiment.

Supramaximal nerve stimulation (using the twitch interpolation method) is a widely used technique used to quantify central and peripheral fatigue (Gandevia et al., 2013; Herbert & Gandevia, 1999; Merton, 1954). To calculate central and peripheral fatigue pre- and post-exercise, electrical impulses were delivered using a nerve stimulator (DS7AH, Digimeter Ltd, UK) during an MVC of the right bicep brachii. The stimulator anode was placed over the medial border of the right scapula, and the cathode was placed on the distal tendon of the right bicep. The anode was then adjusted to achieve the optimal position for stimulation of the biceps brachii, as confirmed based on the force output of the biceps in response to low current stimulation (~50 mA). The stimulation intensity in the main trials was then calculated by progressively increasing the current (10 mA) using increments until a plateau was observed in the mechanical response of the muscle (105 ± 20 mA). All stimulations were delivered using a 300 V, doublet square-wave, with a 10-ms stimulus interval. The location of the carbon rubber electrodes was marked with permanent marker in the familiarization session and replicated for all subsequent experimental trials.

During the post-exercise 60-sec MVC, two stimulations were first given at rest, followed by a 3-sec countdown, at which point participants performed a sustained MVC. Every 10-sec, superimposed twitches were evoked ($Q_{tw,sup}$) over the MVC. The MVC was followed by two more resting potentiated twitches ($Q_{tw,pot}$) exactly 1-sec after full muscle relaxation. Voluntary muscle activation of the bicep (VA%) was calculated using the equation: VA% = (1- $Q_{tw,sup}/Q_{tw,pot}$) x 100 (Folland & Williams, 2007; Lloyd et al., 2015). $Q_{tw,pot}$ was used as an index of post-exercise muscle fatigue. This concluded the physical performance protocol.

Detailed cognitive performance protocol

Immediately following completion of the physical performance protocol, participants were released from the neuromuscular equipment and immediately turned to a standing table to the left side of the neuromuscular rig where they completed the TOH cognitive performance task. The TOH consists of three equidistant pegs which were labelled 1, 2 and 3. There are six different variations of the TOH, i.e., six different starting and ending positions between the three equidistant pegs. Specifically, these variations are from peg 1 to 3, 1 to 2, 3 to 1, 2 to 1, 2 to 3 and 3 to 2. Hence, directly prior to starting the TOH, participants were informed of the variation specific to that condition, and then began the task. Performance ended when the participant successfully moved all discs to the specified end peg.

Physiological and subjective measures

Pre- and post-mental fatigue inducement, as well as pre- and post-physical performance, subjective scales were used to monitor self-reported scores of mental fatigue, mood, sleepiness, and motivation. A 10-point visual analogue scale was used to assess mental fatigue with 0 representing "no fatigue" and 10 representing "worst possible fatigue". Subjective scores of sleepiness using the Stanford Sleepiness Scale (SSS) were also administered to quantify sleepiness (Hoddes et al., 1973). Motivation was monitored using task and success motivation scales (Matthews et al., 2013). Finally, mood was assessed using the Brunel Mood Scale (BRUMS), where subjective scores of confusion, vigour, depression, fatigue, tension and anger were examined (O'Keeffe et al., 2019; Terry et al., 2003).

Throughout the physical performance protocol, subjective ratings of mental effort (Borg CR-100), arm discomfort (Borg CR-10), breathing discomfort (Borg CR-10) and overall exertion (Borg RPE 6–20) were recorded every 3-min (Borg, 1998). Physiological measures, including heart rate (HR), oxygen consumption (VO_2), oxygen saturation (SaO_2), and tissue oxygenation (NIRS) were recorded from the beginning of the 15-min exercise bout. HR was monitored using a Garmin heart rate strap (Model: HRM-DUAL, Garmin, UK) attached around the chest of the participant. VO_2 was recorded using a breath by breath gas analyser system (Model: Quark RMR, COSMED, Italy). Near-infrared spectroscopy (NIRS) (Model: VMS-NIRS, Moor instruments, UK) monitored tissue oxygen of the left bicep brachii and SaO_2 was monitored using a pulse oximeter attached to the left ear lobe (Model: 8500, Nonin, USA).

Data analysis

Normality tests, conducted using the Shapiro-Wilk test, confirmed the data to be both normally and non-normally distributed. The variables which did not conform to normality (i.e., in all conditions) included mood (i.e., confusion, depression, tension and anger), overall motivation and cognitive performance (i.e., time and moves to completion). Both parametric and non-parametric tests were performed, and the data compared for likeness. After comparison, parametric analysis was chosen to perform the data analysis due to the similarity in results and further, due to the extent of the repeated measures and number of variables a more robust and conservative method was deemed more appropriate (Schmider et al., 2010). Hence, a two-way repeated measures analysis of variance (ANOVA) was used for the analysis of the main effects and differential impact of mental fatigue and hypoxia on physical performance and TOH performance (3 x 2: O_2 x MF). A three-way ANOVA including the effect of time was not deemed necessary, due to time yielding a significant interaction over any given time point (Lloyd et al., 2016). Within conditions, pre-and post- were compared using paired sample T-tests with Bonferroni correction for multiple comparisons. A 95% confidence level was used to test

significance ($p < 0.05$). When Mauchly's Test of Sphericity was significant ($p < 0.05$), the Greenhouse-Geisser adjustment was used. This study was sufficiently powered to report a large effect size based on previous research indicating a large effect of mental fatigue on self-paced exercise ($\eta^2_p = 0.70$) (Brownsberger et al., 2013). To calculate the power required to observe a main effect of hypoxia (3 levels), a power analysis conducted in G*Power (Faul et al., 2007) incorporating a large effect size ($f = 0.4$), power of 0.8 with an α err prob of 0.05 yielded a sample size of 10. To calculate the power required for a main effect of mental fatigue (two levels), a large effect size ($f = 0.4$), power of 0.8, with an α err prob of 0.05 yielded a sample size of 12. The power required for a one-tailed (pre to post) t-test using a large effect size of $d = 0.8$, a power of 0.8, and an α err prob of 0.05 outputted a required sample size of 12. Lastly, to calculate the power for the interaction between mental fatigue and hypoxia, PANGEA (Westfall, 2016) an application for power analysis for general ANOVA was used. A large effect size ($d = 0.8$) using 15 participants outputted a power of 0.998. Effect sizes, taken from SPSS, are reported as partial eta squared (η^2_p) for main effects and interactions, and Cohen's d for t-tests. Data are presented as mean ± SD.

Results

Mental fatigue inducement

TloadDback performance

Performance in the TloadDback test remained below 85% accuracy on average in all hypoxia conditions in which mental fatigue was induced (NF, MF, SF), with no difference between conditions, $p \geq 0.187$ (Figure 2). This confirms that participants sustained a high cognitive load in all conditions.

Perceived mental fatigue and sleepiness

There were no significant differences in subjective scores of mental fatigue pre-TloadDback test across the three levels of hypoxia (NF, MF, SF), $p \geq 0.469$. Subjective scores of mental fatigue significantly increased from pre-test to post-test in NF

Table 1. Subjective data quantifying the impact of the mental fatigue test (TloadDback), NF = Normoxia and mental fatigue, MF = Mild hypoxia and mental fatigue, SF = Severe hypoxia and mental fatigue. BRUMS = Brunel Mood Scale. Variables are compared pre- and post-TloadDback. * = significant difference from pre- to post-test scores, $p < 0.05$.

	NF		MF		SF	
Subjective Variable	Pre	Post	Pre	Post	Pre	Post
Mental Fatigue	1.9 ± 1.3	5.7 ± 1.9*	1.7 ± 1.6	6.1 ± 1.8*	1.4 ± 1	4.9 ± 1*
Sleepiness	2.2 ± 0.8	2.6 ± 1.1	2.3 ± 0.9	2.8 ± 1.1	2 ± 0.8	2.5 ± 1
BRUMS: Vigour	33.8 ± 15.3	25.8 ± 16*	29.6 ± 16.6	20.4 ± 21.3	37.5 ± 16	31.7 ± 20.8
BRUMS: Confusion	2.9 ± 7	11.3 ± 8.9*	5.8 ± 9.3	14.2 ± 15.8*	3.3 ± 7.4	12.1 ± 8.7*
BRUMS: Depression	4.6 ± 14.7	5 ± 14.4	4.6 ± 8.3	8.8 ± 13.7	2.5 ± 5.2	4.6 ± 7.3
BRUMS: Fatigue	20.4 ± 12.4	30 ± 11.9	23.8 ± 18.2	39.6 ± 20.1*	14.2 ± 13.7	35.4 ± 13.9*
BRUMS: Tension	7.1 ± 8.1	4.6 ± 8.7	11.3 ± 18.6	11.7 ± 18.3	8.3 ± 16.7	9.2 ± 16.7
BRUMS: Anger	5.8 ± 17.8	6.3 ± 14.2	7.9 ± 15.2	7.9 ± 12.6	1.7 ± 5	2.1 ± 3.9
Motivation: Task	18.2 ± 4.7	15.9 ± 4.8*	17.2 ± 4.6	15.1 ± 4.7*	17.3 ± 5.1	15.2 ± 5.4
Motivation: Success	16.7 ± 6.1	15.9 ± 4.8	17 ± 5.7	16.8 ± 5.6	17.5 ± 4.9	18.1 ± 5.8
Motivation: Overall	2.6 ± 0.8	2.5 ± 1	2.6 ± 0.7	2.3 ± 0.7	2.8 ± 0.9	2.5 ± 1

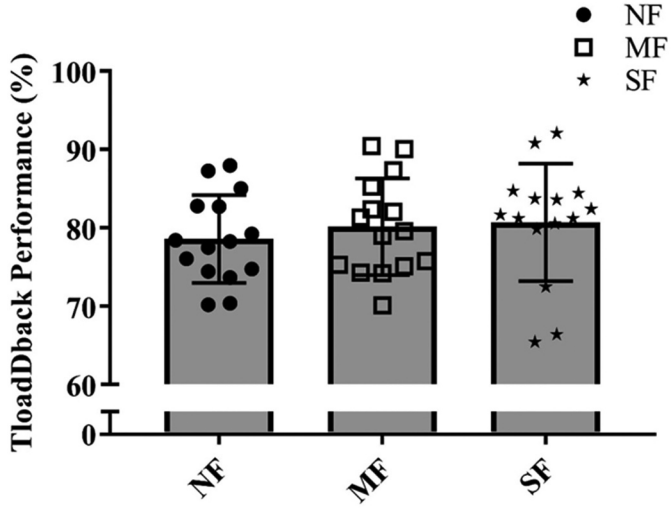


Figure 2. Test Performance in the TloadDback cognitive test for each mental fatigue experimental condition. NF = Normoxia and mental fatigue, MF = Mild hypoxia and mental fatigue, SF = Severe hypoxia and mental fatigue.

($p < 0.001$, $d = -1.88$), LF ($p < 0.001$, $d = -2.84$) and SF ($p < 0.001$, $d = -3.08$) (Table 1). Furthermore, there was no significant difference in subjective mental fatigue across the three levels of hypoxia at post-test, $p \geq 0.207$.

There were no significant differences in pre-sleepiness scores across the three levels of hypoxia, $p \geq 0.334$ (Table 1). Sleepiness scores also did not significantly increase from pre-test to post-test in NF ($p = 0.095$, $d = -0.46$), LF ($p = 0.056$, $d = -0.54$) and SF ($p = 0.068$, $d = -0.51$). Further, sleepiness scores were not significantly different across the three levels of hypoxia at post-test, $p \geq 0.265$. Together, these results suggest that subjective mental fatigue was induced, independent of sleepiness. However, whilst pre-post sleepiness scores did not significantly increase, medium effect sizes were observed. Hence, the lack of significance may be due to low statistical power.

Motivation

There were no significant differences in pre-test task motivation scores, pre-test success motivation scores or pre-test overall motivation scores across the three levels of hypoxia, $p \geq 0.264$ (Table 1). Comparisons between pre-test and post-test task motivation indicated that task motivation significantly decreased in the NF ($p = 0.008$, $d = 0.79$) and MF ($p < 0.001$, $d = 1.21$) conditions, but not in the SF condition ($p = 0.108$, $d = 0.44$) despite reporting a medium effect size, indicating a potential power issue. No significant differences were observed regarding success motivation scores from pre-test to post-test in NF ($p = 0.306$, $d = 0.27$), MF ($p = 0.823$, $d = 0.06$) and SF ($p = 0.541$, $d = -0.16$). Likewise, no significant differences were observed regarding overall motivation scores from pre-test to post-test in NF ($p = 0.751$, $d = 0.08$), MF ($p = 0.055$, $d = 0.54$) and SF ($p = 0.164$, $d = 0.38$). In the MF condition however, given the p-value is close to statistical significance and a medium effect size is observed, the lack of significance may also be due to low statistical power.

Brunel mood scale

No significant differences were observed in pre-test scores of depression ($p \geq 0.075$), tension ($p \geq 0.15$), anger ($p \geq 0.101$), fatigue ($p \geq 0.238$), vigour ($p \geq 0.112$) or confusion ($p \geq 0.138$) across the three levels of hypoxia (Table 1). Comparisons between pre-test and post-test scores of vigour indicated a significant decrease in the NF condition, $p = 0.022$ ($d = 0.66$). No significant differences were found in both the MF ($p = 0.054$, $d = 0.54$) and SF ($p = 0.15$, $d = 0.39$) conditions. In the MF condition the p-value is close to significance and a medium effect size is observed, therefore indicating that the lack of significance may also be due to low statistical power.

Comparisons between pre-test and post-test scores of confusion indicated significant increases from pre-test to post-test in NF ($p = 0.002$, $d = -0.95$), MF ($p = 0.031$, $d = -0.62$) and SF ($p = 0.001$, $d = -1.04$). In addition, comparisons between pre-test fatigue and post-test fatigue scores indicated a significant increase in fatigue in both the MF ($p = 0.001$, $d = -1.05$) and SF ($p = 0.017$, $d = -0.7$) conditions but not the NF condition ($p = 0.058$, $d = -0.533$). However, given the medium effect size, this result could also be related to a statistical power issue. No significant differences were observed in relation to comparisons from pre-test to post-test scores of depression ($p \geq 0.065$), tension ($p \geq 0.305$) or anger ($p \geq 0.751$) across the three levels of hypoxia.

Mental fatigue prior to exercise performance

Subjective scores of mental fatigue were taken pre-TloadDback, post-TloadDback, pre-exercise performance and post-exercise performance to monitor subjective scores of mental fatigue throughout, as presented in Figure 3. Significant differences were found from post-mental fatigue test to pre-exercise, where subjective mental fatigue scores were reduced in NF ($p < 0.001$, $d = 1.18$), MF ($p < 0.001$, $d = 1.23$) and SF ($p = 0.041$, $d = 0.58$). However, significant differences were also found between scores from pre-exercise to post-exercise in NF ($p = 0.012$, $d = -0.74$), MF ($p = 0.033$, $d = -0.61$) and SF ($p = 0.002$, $d = -1.01$), where subjective mental fatigue scores increased across the three levels of hypoxia.

To highlight that subjective mental fatigue scores in NF, MF and SF trials remained significantly higher than baseline scores throughout the testing protocol, comparisons between pre-TloadDback (i.e., baseline) to pre-exercise performance, and between pre-TloadDback to post-exercise performance were also performed. Results indicated that subjective mental

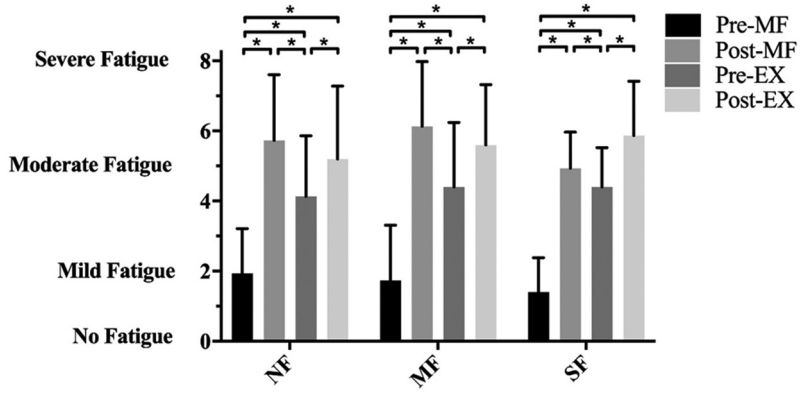
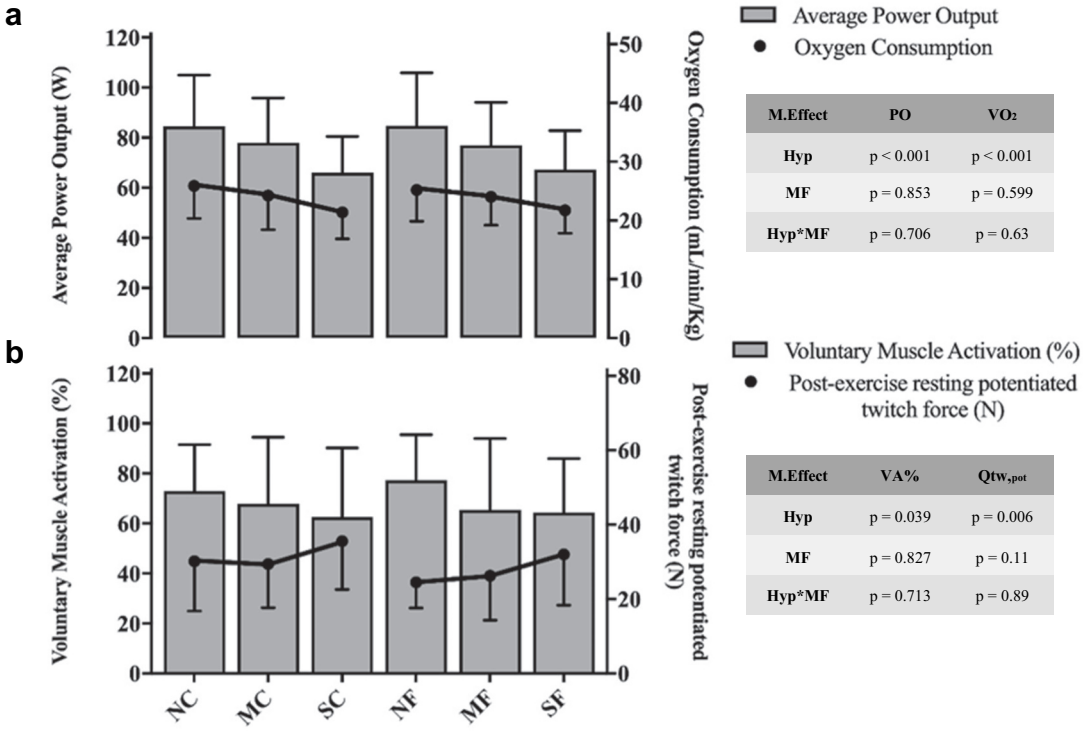


Figure 3. Subjective scores of mental fatigue from pre-TloadDback (Pre-MF), post-TloadDback (Post-MF), pre-exercise performance (Pre-EX) and post-exercise performance (Post-EX), * = significant difference from pre-test to post-test ($p < 0.05$), NF = Normoxia and mental fatigue, MF = Mild hypoxia and mental fatigue, SF = Severe hypoxia and mental fatigue.



M.Effect	PO	VO_2
Hyp	p < 0.001	p < 0.001
MF	p = 0.853	p = 0.599
Hyp*MF	p = 0.706	p = 0.63

M.Effect	VA%	$Qtw_{,pot}$
Hyp	p = 0.039	p = 0.006
MF	p = 0.827	p = 0.11
Hyp*MF	p = 0.713	p = 0.89

Figure 4. A) Average power output (Watts) (left Y-axis) and oxygen consumption (ml/min/kg) (right Y-axis). B) Voluntary muscle activation % (left Y-axis) and post-exercise resting potentiated twitch force (N) (right Y axis) of the biceps brachii. NC = Normoxia control, MC = Mild hypoxia control, SC = Severe hypoxia control, NF = Normoxia and mental fatigue, MF = Mild hypoxia and mental fatigue, SF = Severe hypoxia and mental fatigue. Main effects are presented in the tables to the right of the graphs where M.Effect = main effect, Hyp = hypoxia, MF = mental fatigue, and Hyp*MF = interaction between mental fatigue and hypoxia, PO = power output, VO_2 = oxygen consumption, VA% = voluntary activation percentage, $Qtw_{,pot}$ = post-exercise resting potentiated twitch force.

Table 2. Subjective measures taken during the physical performance protocol in all conditions. NC = Normoxia control, MC = Mild hypoxia control, SC = Severe hypoxia control, NF = Normoxia and mental fatigue, MF = Mild hypoxia and mental fatigue, SF = Severe hypoxia and mental fatigue. ME = Mental Effort measured using Borg CR-100 scale, BD = Breathing Discomfort measured using Borg CR-10 scale, AD = Arm Discomfort using Borg CR-10 scale, RPE = Rating of Perceived Exertion measured using the 6-20 Borg scale. * = main effect of hypoxia, ǂ = main effect of mental fatigue, p < 0.05.

Variable	NC	MC	SC	NF	MF	SF
ME*	39.8 ± 16.5	45.8 ± 20.4	53.9 ± 17.7	44.4 ± 16	47.9 ± 18.8	57.6 ± 14.8
BD*	4.6 ± 1.1	6 ± 1.32	6.5 ± 1	4.6 ± 0.7	5.4 ± 1.4	6.2 ± 1.2
AD*	5.6 ± 0.9	6 ± 1.3	6.5 ± 1.1	5.8 ± 1	6 ± 1.4	6.7 ± 1.3
RPE*	14.4 ± 1.8	14.7 ± 1.8	15.8 ± 1.4	14.7 ± 1.5	15.1 ± 1.4	15.5 ± 1.3

fatigue scores remained significantly higher than baseline scores throughout the duration of the testing protocol, in all hypoxia conditions, p ≤ 0.001.

Physical performance

Power output and oxygen consumption

A main effect of hypoxia was observed on average power output during the time trial, p < 0.001 (η^2_p = 0.79) (Figure 4A). Mental fatigue had no effect on average power output during the time trial, p = 0.853 (η^2_p = 0.003), and no interaction was observed, p = 0.706 (η^2_p = 0.025). Significant differences were seen across all levels of hypoxia, p ≤ 0.002. A main effect of hypoxia was also observed on maximum power output, p < 0.001 (η^2_p = 0.698). No effect of mental fatigue, p = 0.586 (η^2_p = 0.022), or interaction, p = 0.199 (η^2_p = 0.109), was observed on maximum power output.

A main effect of hypoxia was observed on oxygen consumption, p < 0.001 (η^2_p = 0.607) (Figure 4A), however no effect of mental fatigue, p = 0.599 (η^2_p = 0.02), or interaction, p = 0.576 (η^2_p = 0.039), was observed. Near infrared spectroscopy was used to monitor muscle oxygenation. A main effect of hypoxia was observed on muscle oxygen saturation of the bicep, p = 0.004 (η^2_p = 0.327), however no effect of mental fatigue was observed, p = 0.824 (η^2_p = 0.004), and further, no interaction, p = 0.216

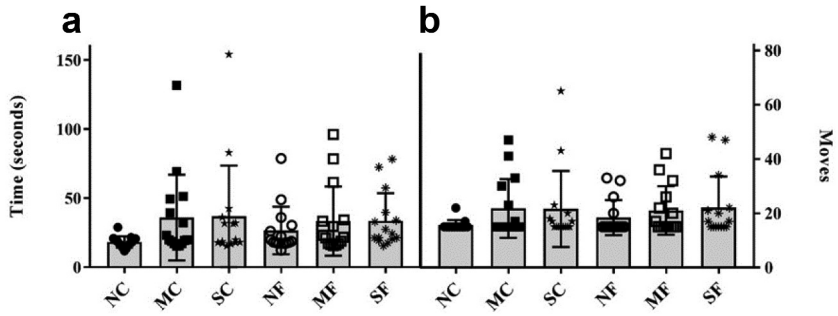


Figure 5. A) Time to completion in the cognitive test, Tower of Hanoi. B) Number of moves to completion in the cognitive test, Tower of Hanoi. NC = Normoxia control, MC = Mild hypoxia control, SC = Severe hypoxia control, NF = Normoxia and mental fatigue, MF = Mild hypoxia and mental fatigue, SF = Severe hypoxia and mental fatigue.

(η^2_p = 0.104). Likewise, a main effect of hypoxia was observed on deoxygenation, p = 0.001 (η^2_p = 0.554), with no effect of mental fatigue, p = 0.281 (η^2_p = 0.082) or interaction observed, p = 0.728 (η^2_p = 0.015).

Central and peripheral fatigue

A main effect of hypoxia was observed on voluntary muscle activation, p = 0.039 (η^2_p = 215) (Figure 4B) while no effect of mental fatigue was indicated, p = 0.827 (η^2_p = 0.004). Furthermore, there was no interaction between hypoxia and mental fatigue on muscle activation, p = 0.713 (η^2_p = 0.017). No significant differences were observed across all six conditions, p ≥ 0.107. A main effect of hypoxia was also observed on post-exercise resting potentiated twitch force, p = 0.006 (η^2_p = 0.305), (Figure 4B) with no effect of mental fatigue, p = 0.11 (η^2_p = 0.172) despite observing a large effect. Further, no interaction was observed, p = 0.89 (η^2_p = 0.008). Together this data indicates that mental fatigue did not decrease the ability of the brain to drive the muscle during maximal voluntary contraction, however in the presence of hypoxia, increases in central fatigue were apparent (Figure 4B).

Subjective measures

Main effects of hypoxia were observed on mental effort, p < 0.001 (η^2_p = 0.664), breathing discomfort, p < 0.001 (η^2_p = 0.75), arm discomfort, p < 0.001 (η^2_p = 0.483) and RPE, p < 0.001 (η^2_p = 0.489) (Table 2). Main effects of mental fatigue were observed on breathing discomfort, p = 0.029 (η^2_p = 0.298). No significant effects of mental fatigue were observed on arm discomfort, p = 0.146 (η^2_p = 0.145), RPE, p = 0.452 (η^2_p = 0.041) or mental effort, p = 0.063 (η^2_p = 0.226). However, given the large effect sizes observed in relation to mental effort and arm discomfort, the lack of significance may be due to low statistical power. No significant interaction between hypoxia and mental fatigue was found in relation to breathing discomfort, arm discomfort, RPE or Mental effort (p ≥ 0.118).

Cognitive performance

Performance in the TOH is indicated by the number of moves and time taken to complete the task. In all six conditions, significant time increases from pre-test performance to post-test performance were observed, p ≤ 0.041. In relation to the amount of time taken to complete the task, no effect of hypoxia (η^2_p = 0.132) or mental fatigue (η^2_p = 0.002) was observed on TOH performance, p≥ 0.138 (Figure 5A). Furthermore, no effect of hypoxia (η^2_p = 0.126) or mental fatigue (η^2_p = 0.013) was observed in relation to the number of moves used, p ≥ 0.152 (Figure 5B). However, given the large effect sizes observed in relation to the effect of hypoxia on cognitive performance, this result may also be due to a lack of statistical power.

Discussion

This study investigated the individual and combined effects of mental fatigue and hypoxia on physical and cognitive performance. In line with previous research, the results from the current study demonstrated that as the severity of hypoxia increased, significant decrements in physical performance were observed. However, in relation to cognitive performance, hypoxia was observed not to decrease TOH performance despite yielding a large effect size. Therefore, the results only partially confirm the first hypothesis. Results demonstrated that mental fatigue was induced in the mental fatigue trials, as indicated by increases in subjective mental fatigue. Further, test performance was sustained below 85% accuracy which is indicative of a high cognitive load required for inducing mental fatigue effectively using this test (Borragán et al., 2017). Sleepiness and overall motivation were not significantly influenced by the mental fatigue protocol despite reporting medium effect sizes, indicating a potential lack of statistical power. Nevertheless, the results of the current study indicate that physiological arousal was maintained prior to physical performance. Despite mental fatigue being induced and sustained however, the results observed no effect of mental fatigue on either physical or

TOH performance, hence rejecting both the second and third hypotheses.

Physical performance

In line with previous research, the current study confirmed that hypoxia decreased physical performance, where average power output and oxygen consumption were both reduced as the severity of hypoxia increased (Lahiri et al., 1976; Fulco et al., 1998; Wehrlin & Hallén, 2006; Bhaumik et al., 2008; Calbet et al. 2002). Similarly, a reduction in voluntary activation of the bicep brachii was observed on exposure to increasing hypoxia, indicating an increase in central fatigue. In normoxia and mild hypoxia, the results indicate similar end-exercise peripheral fatigue, suggestive of a potential critical threshold of peripheral fatigue protecting the exercising limb. Therefore, the results suggest that voluntary activation is reduced via feedback from the muscles' sensory neurons (i.e., group III/IV muscle afferents) restricting motoneuronal output and therefore voluntary activation in proportion to the magnitude of the feedback (Goodall et al., 2012; Hureau et al., 2016).

Furthermore, an increase in post-exercise resting potentiated twitch force was observed indicating an increase in end-exercise peripheral fatigue relative to the increase in severity of hypoxia. This was expected due to the decline in neural drive, as indicated by the decline in VA%. Therefore, during self-paced exercise in severe hypoxia, it is evident that the reduction in neural drive (i.e., central fatigue) is necessary to compensate for the increasing fatigued motor units (i.e., peripheral fatigue). Previous research investigating neuromuscular fatigue in the presence of hypoxia support these findings whereby hypoxia facilitates the onset of central and rates of peripheral fatigue development (Goodall et al., 2010; Herbert & Gandevia, 1999; Lloyd et al., 2016).

The current study observed no effect of mental fatigue on physical performance, despite mental fatigue being induced and maintained prior to physical performance. Previous research reporting the negative impact of mental fatigue on physical performance has used single-task tests that are of long durations (>30-min) (Marcora et al., 2009; Van Cutsem et al., 2017), and are not individualized to each person's individual cognitive processing speed (O'Keeffe et al., 2019). Hence, in previous research, it is unclear whether the performance decrements observed are due to mental fatigue or states of sleepiness or under-arousal. In the current study, however, mental fatigue was induced prior to physical performance, using an individualized and short-duration cognitive test, independent of subjective sleepiness and whilst maintaining arousal (O'Keeffe et al., 2019). Therefore, the results suggest that, mental fatigue per se, had no effect on physical performance. Future research investigating the impact of mental fatigue, independently of sleepiness, on physical performance is perhaps warranted.

Perceived exertion and discomfort

There is, however, research which similarly has found no effect of mental fatigue on physical performance (Holgado et al., 2020; Pageaux et al., 2013; Vrijkotte et al., 2018). Holgado et al. (2020) replicated the seminal study by Marcora et al. (2009) to investigate the reliability of their findings, which demonstrated that mentally fatigued subjects (induced using 90-min of the AX-CPT task) reached exhaustion (i.e., cycling time to exhaustion protocol at 80% peak power output) faster than non-mentally fatigued subjects. They found that although mental fatigue was subjectively attained (albeit without controlling for sleepiness or arousal), mental fatigue did not impact physical performance (Holgado et al., 2020).

Marcora et al. (2009) further hypothesized that the negative impact of mental fatigue observed on physical performance was driven by reductions in RPE. Hence, their findings suggested that a greater level of subjective mental fatigue would lead to a higher RPE, and hence would subsequently impair performance, a finding supported by follow up studies (Pageaux et al., 2014; Van Cutsem et al., 2017). Conversely, in the replication study (Holgado et al., 2020), no increase in RPE was observed when participants were mentally fatigued compared to control. However, a limitation of this study is that Holgado et al. (2020) measured session RPE as opposed to RPE over the course of the trial as in previous research. Nevertheless, the current study monitored RPE over the course of the physical performance protocol (i.e., every 3-min) and found only an impact of hypoxia on RPE, with mental fatigue having no effect. In addition, hypoxia increased ratings of mental effort, breathing discomfort, and arm discomfort, which may have further contributed to the reduced physical performance observed. Given these results, research is warranted to investigate the potential use of psychological skills interventions in hypoxia, where improved exercise performance may be possible through mitigating subjective discomfort and exertion.

Cognitive performance

In the current study, the results displayed no main effect and no interaction between mental fatigue and hypoxia on TOH performance. No effect of hypoxia was observed on cognitive performance despite results indicating a large effect size. Therefore, the null effect of hypoxia observed in the present study could be due to a lack of power. Contrasting to the results of the current study, previous research investigating the impact of both acute and chronic hypoxic exposure on cognitive performance has observed a negative impact of hypoxia on cognitive processing (Abraini et al., 1998; Griva et al., 2017; Kramer et al., 1993; McMorris et al., 2017; Taylor et al., 2016). This decline in cognitive performance is a consequence of both hypoxaemia, imposing a reduction in oxygen saturation in arterial blood, and the resulting hypocapnic response, a reduction in the arterial pressure of carbon dioxide. However, previous research has also found no effect of hypoxia on cognitive performance due to mechanisms such as behavioural regulation strategies (Lefferts et al., 2019), the complexity of the cognitive task (i.e., simple tasks v complex tasks), duration of exposure to hypoxia (Martin et al., 2019; Williams et al., 2019), the post-acute supramaximal exercise oxyhaemoglobin response (Bediz et al., 2016), issues such as poor methodological design and the disparate use of cognitive tests (Williams et al., 2019), and the intertwined balance between the beneficial effects of acute exercise and detrimental effects of

severe hypoxia (Komiyama et al., 2017). In the current study however, we did not measure such mechanisms to help explain the null effect of mental fatigue and hypoxia on cognitive performance, thus, we can only speculate the mechanisms underpinning the performance outcome. In addition, previous research has presented a negative impact of mental fatigue on cognitive performance (Holtzer et al., 2011; Tanaka, 2015). However, despite mental fatigue scores significantly increasing from pre-exercise to post-exercise (prior to the TOH) no effect was observed.

Williams et al. (2019) recently examined the physiological mechanisms underpinning cognitive performance at rest, in different levels of hypoxia. They found that performance in a complex cognitive task (i.e., n-back test) was reduced at FiO_2 0.12 due to reductions in cerebral oxygenation and peripheral oxygen saturation. The TOH is also classified as a complex cognitive performance task which has been shown to indicate prefrontal lobe function and dysfunction and executive processes such as working memory, inhibition and fluid intelligence (Welsh & Huizinga, 2005). Williams et al. (2019) proposed that the reduced performance in the complex cognitive task could also be a result of the task requiring the activation of different brain regions in order to successfully perform the task, hence making it more susceptible to reductions in performance due to hypoxia. Contrasting to this finding, no impact of hypoxia was observed on performance in the TOH, also a complex task, with the only difference being that the cognitive task was performed post-exercise performance. Hence, the beneficial post-exercise facilitation of brain oxygenation could explain the null effect of hypoxia on cognitive performance. However, this suggestion requires further research since measures such as cerebral oxygenation and cerebral blood flow to monitor this post-exercise response on cognitive performance in hypoxia were not taken in this study.

Neuropsychological mechanisms

Large interindividual variability in cognitive performance (Martin et al., 2019; Williams et al., 2019) and physical performance (Fulco et al., 1998) has been observed in hypoxia. However, limited research has attempted to rationalize this variability considering trait psychological constructs. Such psychological constructs that have been shown to enhance and predict physical performance include emotional intelligence (Kopp & Jekauc, 2018) and mental toughness (Gucciardi et al., 2015). People who possess higher levels of emotional intelligence, described as the understanding and awareness of and ability to regulate emotions (Laborde et al., 2016), have demonstrated a greater tolerance to physiological stress without impeding performance. Further, people that have greater levels of mental toughness, a unidimensional construct encompassing values, attitudes, emotions and cognitions (Swann et al., 2016), can sustain high levels of subjective and objective performance despite impeding challenges, through enhanced perception of stress and more effective coping strategies (Gucciardi et al., 2015). Considering these trait psychological constructs and their relative impacts on performance, it is possible that the interindividual variability and the null effect of mental fatigue could be due to individuals in the present study possessing such trait psychological characteristics and hence utilize effective coping mechanisms to prioritize the focus of attention and block out some sensations of pain and fatigue. Future psychophysiological research should aim to measure trait psychological constructs such as emotional intelligence and mental toughness which may aid in explaining the potential variability in performance.

A further rationale to why mental fatigue did not impact physical performance could be due to the interconnection between the underlying neural mechanisms of mental and physical fatigue. Neuropsychological research has reported that, as active muscle fibres become fatigued through exercise, the primary motor cortex is increasingly stimulated to compensate for muscle fatigue (Tanaka et al., 2014, 2014; Lloyd et al., 2016). This process is known as the physical facilitation process and is regulated by neural networks which overlap with the neural networks involved in the "mental facilitation" process which aids in combatting mental fatigue (Tanaka et al., 2014). Hence, when an individual is tasked with combatting both mental and physical fatigue simultaneously, it could be hypothesized that the worst stressor, i.e., physical fatigue or mental fatigue, and their corresponding neural facilitation networks, will take precedence (Lloyd & Havenith, 2016; Lloyd et al., 2016). Results from the current study suggest that the physical exertion component of the trial (i.e., 15-min self-paced exercise and maximal voluntary contraction) may have overridden the impact of mental fatigue through underlying neural mechanisms. Future research would benefit from the inclusion of measures to report the impact of such neuropsychological factors.

Limitations

A potential limitation to this study was the transition period between completing the mental fatigue test (outside the environmental chamber) and conducting the physical performance protocol (inside the environmental chamber) as this transition period may potentially have reduced the impact of mental fatigue. To reduce the time of this transition, participants were instructed to immediately mount the rig on entry to the environmental chamber, all equipment was attached, and the time trial started in less than 5-min. To monitor the maintenance of mental fatigue throughout the study, subjective scores were recorded at four different times. Compared to baseline ratings of mental fatigue (i.e., pre – TloadDback test), throughout the duration of the trial (i.e., post-TloadDback test, pre-exercise performance, post-exercise performance), scores of mental fatigue were significantly higher than baseline scores. Hence, we can confirm that subjective scores of mental fatigue were maintained throughout the experimental protocol, and that the transition time did not impact the maintenance of mental fatigue.

This study utilized an arm-bike ergometer, which was a relatively unfamiliar task to all participants. However, despite its unfamiliarity, this mode of exercise is not difficult and can be undertaken effectively. After turning a number of efficient forces, it simply requires a pushing effort with one arm and a pulling effort with the other. A familiarization session included a full run through of the hand bike ergometer protocol so that participants could practice this

mode of exercise performance prior to beginning the main experimental trials.

As the current study aimed to investigate whether mental fatigue and hypoxia both independently and combined would impact physical and cognitive performance, the mental fatigue test was conducted in pre-exercise normoxia. This provides scope for further research to investigate whether performance would be impacted when mental fatigue is induced in a hypoxic environment. Likewise, it would be interesting to explore how the individual stimulus times would be affected if participants were required to achieve these in a familiarization session conducted in a hypoxic environment.

The current study would have benefited from the incorporation of objective brain activity measures such as Functional Near-Infrared Spectroscopy (fNIRS) and Electroencephalography (EEG). By using these methods, we would be able to objectively quantify the impact of mental fatigue and monitor its maintenance, increase or decrease throughout the experiment. Further, by observing brain activity during performance in the TOH, we would see the relative performance in normoxia compared to hypoxia, and further evaluate the post-exercise oxy-haemoglobin response on performance in the TOH.

Lastly, it is important to acknowledge the limited power of the study which may explain why some of the results have reported large effect sizes yet are not statistically significant. In particular, where significance values are close to $p = 0.05$ and variables output large effect sizes (i.e., $\eta^2 \geq 0.14$), a higher-powered study may yield different results.

Conclusion

This study is the first to investigate the impact of mental fatigue on physical and cognitive performance in hypoxia. A unique element of the research involved participants completing a 16-min dual-task cognitive test that was individualized to participants' cognitive processing speeds. The results indicate that mental fatigue was induced, independently of sleepiness, prior to performance in the physical performance test. Despite mental fatigue being induced however, the results confirm that it had no impact on either physical or cognitive performance in normoxia or hypoxia. Potential rationales as to why mental fatigue did not impact physical or cognitive performance could be due to interindividual variability in psychological constructs such as emotional intelligence and mental toughness, and the possible impact of the physical stressor, i.e., physical exertion, overriding the impact of mental fatigue. This research provides preliminary evidence that mental fatigue, independent of sleepiness, is a transient phenomenon. Overall, physical performance was negatively impacted by hypoxia. Further, cognitive performance in the Tower of Hanoi was not impacted by hypoxia, potentially explained through the exercise-hypoxia interrelationship in maintaining cerebral oxygenation, behavioural regulation strategies in hypoxia to sustain accuracy and interindividual cognitive processing abilities.

Disclosure statement

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Author contributions

KO, AL, and SH conceived and designed the research. KO and GR conducted the experiment. KO analysed the data with support from AL. KO drafted the manuscript. All authors read, revised critically and approved the manuscript.

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