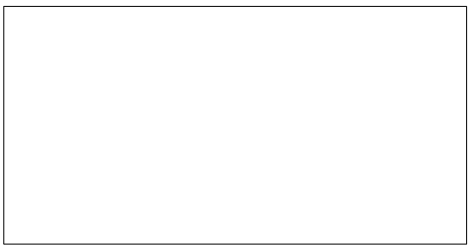

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Elevated Arousal following Acute Ammonia Inhalation is not Associated with Increased Neuromuscular Performance

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Brief running head: Ammonia Inhalation, Arousal and Neuromuscular Performance

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Abstract

Many athletes seek to enhance their performance using legal ergogenic aids, including ammonia inhalants (AIs). AIs trigger the inhalation reflex and increase blood pressure, respiration and heart rate; but, despite their widespread use, there is little evidence for the benefits of AI on exercise performance. We aimed to determine the psychological and neuromuscular impact of acute ammonia inhalation. Fourteen non-resistance trained males completed three trials: control, experimental (AI), and sham. The order of the sham and experimental trials was randomized. Participants completed handgrip and knee extension maximal voluntary contractions (MVC), and countermovement jump (CMJ). Heart rate and alertness were recorded at rest and immediately following control, experimental or sham treatment, followed by functional performance measurements. Reaction time, electromechanical delay, rate of force development and peak force were calculated from MVCs, and peak power from CMJ. On completion of trials, perceived performance was recorded. Statistical significance was accepted at $P<0.05$. Heart rate ($P<0.001$), alertness ($P=0.009$) and perceived performance ($P=0.036$) were elevated by AIs. Markers of functional performance were unaltered by AIs. Alertness was moderately correlated with perceived performance in control ($r=0.61$) and sham conditions ($r=0.54$), and very-highly correlated in the experimental condition ($r=0.90$). AI elevates alertness and perceived physical performance, but not peak strength, power, or neuromuscular drive. AIs may be a useful psychological stimulant to increase focus and mental preparation, however it is unlikely that this will improve functional performance in an untrained population. Our data suggest however, that ammonia inhalants may improve the perception of an individual's performance.

Keywords: Smelling Salts, Ergogenic Aid, Stimulant, Alertness, Performance

Introduction

Muscle force and power are key determinants of physical health and sporting performance (1, 2). While chronic adaptations in such functional components are influenced by factors including muscle mass, morphology, and neural activation (3); psychological arousal may also have an acute role in influencing functional performance (4). As such, maximising both physical and psychological capacity will enhance performance (5). While mass and morphology can only effectively be enhanced through continual exercise training, neural activation and arousal can be altered instantaneously (6). Because of the potential instantaneous performance benefits associated with enhanced arousal state (4) many athletes use ergogenic aids in pursuit of superior performance during competition (7). For example, various ergogenic aids have been shown to increase central nervous system (CNS) activation, improving performance outcomes (7). While some such aids are banned within sport because of potential health risks (8), there remain many legal aids, but most of these lack rigorous evidence to support their application (9, 10).

Legal supplementation is commonplace throughout competitive sport. In a survey of American athletes in the collegiate system, 89% reported using nutritional supplements (11), with ~50% of powerlifters reportedly making use of non-nutritional ergogenic aids during competition, including ammonia inhalants (AIs) (12). AIs are a common example of stimulant-based ergogenic aid used within competitive sport; yet, despite their popularity, there has been little research into their effect on sporting performance. Greater anaerobic power has been observed with AI use in a fatigued state (13), but they have been shown not to impact lower limb or full body static strength (14, 15), dynamic strength (16, 17), or number of repetitions to fatigue (16). However, no previous study has assessed the influence of AIs on isolated upper body

strength only. Meanwhile, rate of force development (RFD) may (14) or may not (15) be influenced by AI use. While the above-mentioned findings all relate to experienced resistance- or anaerobically-trained individuals, it has been proposed that untrained individuals may benefit from psychological arousal to a greater extent than those who are trained (4). Arousal has been widely assessed by measuring alertness (18, 19); however, to date no study has assessed alertness specifically following AI use.

Despite the scarcity of evidence for their efficacy, AI use remains widely popular among those competing in sports such as powerlifting, weightlifting, track and field, boxing, American football, hockey and mixed martial arts (20). To help understand their popularity, we can explore the physiological responses induced by AI use. It is believed that ammonia inhalation triggers the trigeminal nerve via chemoreceptors within the nasal, oral and pulmonary mucosa (21). The respiratory and vasomotor centres within the medulla oblongata respond to this irritation, promoting inhalation reflex and increased blood pressure leading to elevated respiration and heart rate (HR) (22). Perry et al. (2016) evidenced this response, demonstrating increased middle cerebral artery blood flow velocity and HR immediately following AI use (15). Furthermore, improved cerebral delivery of oxygenated blood and CNS excitation, result in increased consciousness and enhanced sympathetic activity. In terms of an advantageous effect for sporting performance, these responses could potentially elicit cognitive enhancement and increase central drive (13). Equivocal findings of an effect of AIs on RFD (14, 15) compel further investigation; if there is, as suggested, an increase in central drive associated with AI use, elevated RFD would be expected, accompanied by a shortening of electromechanical delay (EMD), and therefore, overall reaction time (RT). These enhancements could be expected to promote an increase in peak power production, although to date the only study to report an AI-

induced increase in peak power, investigated performance in already-fatigued athletes (13). Therefore, it remains unclear whether AIs can acutely enhance neuromuscular processes sufficiently to impact functional performance.

The aim of this study was to determine the effect of AI use on peak power production, and on RT and RFD during maximal isometric strength assessment, among non-resistance trained individuals. Additionally, we assessed the effect of AI use on HR and cognitive alertness and examined the relationship between these parameters and functional performance. As arousal may acutely benefit performance and has a greater impact on untrained than trained individuals, we believe that studying an exclusively untrained cohort may provide the best opportunity to explain the popularity of AI use via its effect on arousal and enhanced psychological readiness to perform. Furthermore, enhanced arousal and subsequent psychological readiness to perform may translate to an overall perception of improved performance compared to when not using the stimulant and thus, may potentially help to further explain the popularity of AI use. We therefore hypothesised that alertness would increase in line with HR following AI use, and that this would be associated with decreased RT and increased RFD, which would result in elevated peak power, but not peak strength. It was also hypothesised that participants would perceive a sense of improved overall performance after AI use.

Methods

Participants

Fourteen non-resistance trained male participants with no history of neuromuscular or musculoskeletal disorders were recruited (age 20 (SD 1) years, height 179.9 (SD 5.4) cm, body

mass 76.2 (SD 12.7) kg, weekly physical activity 3.5 (SD 1) days). Participants were required to read a study information sheet and complete a pre-participation questionnaire to determine eligibility. Eligible volunteers provided written informed consent after being supplied with information about any benefits and potential risks involved with their participation. The study was approved by the local Research Ethics Committee (SSREC No. 701/705), and all procedures performed were in accordance with the ethical standards of the 1964 Helsinki declaration and its later amendments. Participants were eligible if they were over the age of 18, had not previously used AIs, and had no known respiratory or cardiovascular illnesses.

Design

Following full familiarisation of the testing procedures, participants completed 3 trials separated by 7-days, each at the same time of day. The three trials consisted of control (CN), experimental (AI) and sham conditions, where the order of the latter two was chosen by participant block randomisation. Trial 1 (CN) also acted as a participant familiarisation and followed an identical protocol to trials 2 and 3, except that no inhalant was provided (Figure 1). During trials 2 and 3 participants inhaled either AI within the experimental condition or water within the sham condition prior to all measurements of functional performance. A sham condition of water was chosen over a scented alternative (as has been adopted elsewhere (16)) as evidence shows that other strong smelling substances can promote muscle activity (23). After the control trial, participants were informed that they would inhale the AI during one trial, and water (sham) during the other, and that these trials would be randomised in order. The order of the AI and sham trials was not revealed to participants prior to inhalation. Functional performance was determined via knee extension (KE) and handgrip (HG) maximal voluntary isometric contraction (MVC) using dominant limbs, and bilateral countermovement

jump (CMJ); each incorporating 3 repetitions. Each participant identified their dominant upper and lower limbs by indicating their handedness, and the leg they would use to kick a ball (24), respectively. Participants rested for 20-minutes between tasks, and 5-minutes between repetitions. Finally, participants self-rated their perceived performance across the 3 tasks (KE and HG MVCs, and CMJ). The order of measurements was consistent across all visits (Figure 1).

[Figure 1]

Procedures

Participants refrained from strenuous physical activity, alcohol, and caffeine for ≥24h preceding laboratory visits, and maintained similar diet for 48h, incorporating 1h fast immediately prior to each trial. Upon arrival to the laboratory, participant height and mass were recorded before resting in a semi-reclined, supine position for 20-minutes. Next, resting HR was recorded (FT1 Polar monitor, FT7 sensor. Warwick, UK); three 5s stable readings were noted, and the mean value was recorded for analysis; intraclass correlation coefficient (ICC) of resting HR between trials 1, 2 and 3 was 0.85 with no significant differences in resting HR between trials. Participants then indicated their alertness on a 200mm visual analogue scale (VAS), which ranged from 'not alert' at the extreme left to 'highest possible alertness' at the extreme right, which is believed to be similar to an alertness VAS which have been used previously (25, 26). The two ends of the VAS were anchored by perpendicular lines, but there were no increments between the end markers. This method was chosen as VAS has been shown to be a reliable and valid assessment of alertness (25, 26). The ICC of pre-inhalation alertness

between trials 1-3 was 0.94, and there were no significant pre-inhalation differences in alertness between trials ($P>0.05$). During trials 2 and 3, participants inhaled from an unmarked flask containing either 35.99g of AI (Nose Tork, Crystals of Ammonium Carbonate, Oklahoma, US) or water and cotton wool before all MVC and CMJ repetitions. Both flasks were matched exactly in volume and appearance, although the scent and physical reaction of participants after inhalation could not be controlled for between the experimental and sham trials. Participants were instructed to inhale sharply, while the flask was positioned ~150mm below the columella. HR and alertness were recorded as before, immediately following inhalation prior to one of the three repetitions, allocated at random. During CN (trial 1), participants followed the same procedure, but no flask was positioned below the columella.

For assessment of MVC, participants were coupled to an isokinetic dynamometer (Biodex System 3, Medical Systems, Shirley, New York, USA). In accordance with the manufacturers' instructions, seat positions were adjusted to suit each participant's anthropometric characteristics, and straps (across the chest and pelvis) were used to secure the participant in the required position. The final positioning of each participant was recorded on the initial visit and replicated throughout the experimental period to ensure constancy between trials. Two pairs of Ag/AgCl self-adhesive electrodes (PNS Dual Element Electrode, Vermed, Vermont, USA) were affixed to pre-prepared skin, 1 pair over vastus lateralis and 1 pair over brachioradialis, in accordance with SENIAM guidelines. Ground electrodes were affixed over patella and olecranon, respectively. Surface electromyogram (sEMG) was captured during all MVCs, at a sampling rate of 2kHz, via Acknowledge® 3.9.1.6 software integrated with Biopac MP100 hardware (Biopac Systems Inc., Goleta, California, USA). For KE MVC a Velcro cuff was secured proximal to the medial malleolus. Gravitational corrections were performed, in

accordance with existing recommendations, in order to account for the effect of limb weight. The lateral femoral epicondyle of the dominant leg was visually aligned with the dynamometer axis of rotation, and a knee joint angle of 60° was set (0° = full extension) (27). Participants performed a standardised submaximal warm-up, consisting 2 sets of 3 x 5s isometric KE contractions at 50% and 75% of perceived maximum, respectively. During all contractions participants were instructed to cross their arms in front of their chest. For HG MVC the shoulder of the dominant arm was flexed to 90° (0° = neutral), and secured to a support, with an elbow joint angle of 90° (28). A similar warm-up protocol was performed, using a HG dynamometer (MLT003/D, AD Instruments Inc. Colorado Springs, Colorado, USA), and the contralateral arm remained in a neutral position.

Following the respective warm-ups, MVC was assessed via 3 x 5s maximal contractions. During trials 2 and 3 participants inhaled either AI or water prior to each contraction. The time between inhalation and contraction varied by small durations between each test due to the randomised audio prompt which was used to measure RT. However, time between inhalation and contraction did not exceed 30 seconds. Dynamometer outputs were sampled synchronously with sEMG via Acknowledge® 3.9.1.6 (Biopac Systems Inc., Goleta, California, USA). Participants were instructed not to hold back any effort for subsequent contractions; the same investigator provided standardized verbal commands and encouragement, to assist the participants in achieving maximal effort for every contraction. Participants reacted to an audio prompt and were instructed to exert as much force as possible, as quickly as possible, in response to the prompt, allowing RT and RFD to be calculated (29). The contraction containing the highest peak was designated MVC. The ICC for HG and lower limb MVCs between trials was 0.96 and 0.94, respectively. From these contractions, RT

(latency between audio stimulus and onset of force production) and EMD (latency between onset of sEMG and onset of force production) were calculated. ICCs for RT and EMD were calculated as 0.81 and 0.13 for HG, and 0.88 and 0.14 for the lower limb, respectively. Additionally, RFD was analysed over 0-20, 0-50, 0-100 and 0-200ms from the onset of force production. In all cases, onsets were considered as $\geq$2SD from baseline. ICCs between trials for HG and lower limb, respectively, are as follows: 0-20ms (0.63 and 0.58), 0-50ms (0.74 and 0.81), 0-100ms (0.75 and 0.88), and 0-200ms (0.77 and 0.94). Analysis was performed using MATLAB version 7.11.0.584 (R2010b) software (The MathWorks, Inc. Natick, Massachusetts, USA) and force and sEMG onsets were visually confirmed *post hoc* (29).

Peak power was assessed via CMJ (JUMP-MD, Vertical Jump Meter T.K.K.5406) according to the formula: $W = (78.6 * d) + (60.3 * m) - (15.3 * h) - 1308$ (where: W = peak power, d = displacement, m = body mass, h = height) (30). First, participants warmed-up by completing 3 CMJs at 50% of perceived maximum effort. Three maximal effort CMJs were then performed, with 5 minutes rest between attempts; during trials 2 and 3 participants inhaled either AI or water prior to each attempt. The greatest displacement achieved across the 3 attempts was used to calculate peak power. The ICC of all trials was calculated as 0.97. Following assessment of all functional measures, participants self-rated their perceived performance level using a 200mm VAS which ranged from 'worst performance' at the extreme left to 'best possible performance' at the extreme right (31). The two ends of the VAS were anchored by perpendicular lines, but there were no increments between the end markers. Perception of performance ICC between the three trials was calculated as 0.85.

Statistical Analyses

Data were assessed for normal distribution and equal variances using Shapiro-Wilk and Levene's Test for Homogeneity of Variances, respectively. The percentage change (Δ%) in pre- to post-inhalation HR and alertness was firstly calculated for all three conditions. These data were subsequently used to calculate the percentage change from the CN during the AI and Sham conditions. MVCs, CMJ power, EMD, RT and perceived overall performance were also calculated as the percentage change from the CN. Heart rate, alertness, MVCs, CMJ, EMD, RT and perceived overall performance were then analysed using a paired sample Student's *t*-test to identify significant differences between AI and sham conditions. Wilcoxon W rank test was used when normality assumptions were violated. Cohen's D (d) effect size was subsequently calculated for t-tests, where values of 0.2, 0.5 and 0.8 represented a small, medium and large effect, respectively. Additionally, RFD (0-20, 0-50, 0-100 and 0-200ms) from each condition were analysed using two-way repeated measures ANOVA. Where significant and non-significant effects were observed, partial η^2 effect sizes (η^2_p) were calculated by $\eta^2_p = SS_{conditions} / (SS_{conditions} + SS_{error})$. Association between pairs of variables was assessed using Pearson's correlation coefficient. Correlation coefficients were interpreted as < 0.30 = *negligible*, 0.30 - 0.49 = *low*, 0.50 - 0.69 = *moderate*, 0.70 - 0.89 = *high*, 0.90 - 1.00 = *very high* (32). Statistical significance was accepted at $P<0.05$. All statistical analysis was performed using Jamovi (Version 0.9), and figures were created using Microsoft Excel (Version 16.31) and Jamovi.

Results

When measured relative to the CN condition, the percentage difference in heart rate ($t_{(13)}$=4.76, d=1.27, P<0.001) and alertness ($t_{(13)}$=3.1, d=0.837, P=0.009) were elevated by 11.5% [95% CI: 6.3, 16.7] and 20.1% [6.1, 34.2] respectively following AI use compared to the sham condition (Figure 2). The change in perception of performance from the CN condition was also higher after AI use by 8.6% [0.66, 16.6] when compared to the sham condition ($t_{(13)}$=2.34, d=0.63, P=0.036; Figure 2C).

[Figure 2]

KE MVC ($W_{(13)}$=72, , *d=0.31*, P=0.24), HG MVC ($t_{(13)}$=-0.264, d=-0.07, P=0.80), and peak power ($t_{(13)}$=0.625, d=0.17, P=0.54) were all unaffected by AI use. The percentage change in RT and EMD from the CN also did not differ between conditions during KE MVC ($t_{(13)}$=1.72, d =0.46, P=0.11; $t_{(13)}$=0.13, d=0.03, P=0.9, respectively), or during HG MVC ($W_{(13)}$=46, d=0.08, P=0.72; $t_{(13)}$=1.34, d=0.36, P=0.2, respectively) (Table 1). RFD during KE MVC ($F_{(2,13)}$=3.1, η^2_p=0.04, P=0.064) and HG MVC ($F_{(2,13)}$=3.27, η^2_p=0.2, P=0.54) did not differ between the three conditions. However, a significant main time effect for RFD development between 20 – 200ms was observed during the KE and HG MVC ($F_{(2,13)}$=9.7, η^2_p=0.08, P<0.001; $F_{(2,13)}$=50.2, η^2_p=0.79, P<0.001, respectively), and the results of post-hoc analysis are displayed in Table 1).

[Table 1]

There were positive correlations between alertness and perceived performance post-AI ($r_{(12)}$=0.90, P<0.001), post-sham ($r_{(12)}$=0.54, P=0.046), and post-CN ($r_{(12)}$=0.61, P=0.02) (Figure 3). There was no relationship between HR and alertness post-AI ($r_{(12)}$=-0.33, P=0.25), post-sham ($r_{(12)}$=-0.20, P=0.50), or post-CN ($r_{(12)}$=0.07, P=0.81); nor between HR and perceived performance post-AI ($r_{(12)}$=-0.25, P=0.38), post-sham ($r_{(12)}$=-0.15, P=0.62), or post-CN ($r_{(12)}$=0.06, P=0.84).

[Figure 3]

Discussion

The aim of this study was to examine the effects of ammonia inhalant use on functional performance and psychological alertness, in non-resistance trained males. Despite being commonly used as a performance-enhancing stimulant, we observed no improvement in any aspect of functional performance following AI use. Interestingly however, use of the stimulant did elicit an increase in individual perception of overall performance, which was very highly associated with elevated rating of alertness.

Despite a lack of research existing to support their beneficial effects, AIs are commonly used in resistance-based exercise as they are believed to temporarily increase consciousness and arousal of an individual during training and competition (20). The current study is the first to

assess arousal, via an alertness scale, alongside markers of functional performance, following AI use; specifically, we observed an increase in alertness ratings after stimulant inhalation. It is possible that alertness was elevated as a result of a sympathetic nervous response, as AIs have previously been reported to increase breathing rate and HR following inhalation (15, 20, 21). However, despite an acute elevation in HR (Figure 2A), there was no correlation between HR and alertness, which suggests that sympathetic nervous response was not exclusively responsible for increases in alertness with AI use. Elsewhere, Perry et al. (15) evidenced an acute increase in blood flow to the brain, as measured by middle cerebral artery blood velocity, immediately after AI use. Consequently, our finding that alertness was elevated following AI use may be attributable to greater delivery of oxygenated blood to the brain (33). It is known that a reduction in cerebral blood flow impairs cognitive function (34), which may well be linked to performance decrements associated with inadequate levels of arousal during training and competition (35). With this in mind, AI-induced enhancement of alertness may help to explain the popularity of this particular stimulant within performance sport. However, such interpretations are beyond the scope of this study.

The current study was also the first to investigate the effects of AIs on perception of overall performance, with participants self-rating that a superior performance was achieved during the AI trial (Figure 2C). Interestingly, moderate-very high positive correlations were observed between perceived performance and alertness ratings within all three conditions, with the highest correlation observed in the AI trial (Figure 3). It is possible that elevated alertness induced by AI use could have improved participant's perception of their performance by enhancing focus and ensuring optimal psychological state was achieved. Furthermore, AIs are believed to increase not only the focus, but also the effort exerted by athletes (20). Subsequently, an individual's reflection of their performance with increased alertness, reduced

distraction, and potentially higher levels of effort may have translated into higher self-rating of perceived performance. An improved psychological readiness to perform and consequently, enhanced perception of performance may provide further evidence as to why AI use is prevalent during competition (11, 12). Furthermore, limited data exist regarding the use of AI within different sporting performances. Therefore, when uninformed on physical performance, individuals may argue that AI use are important as from a perceived performance perspective, they believe to perform better after using the stimulant.

Although psychological factors were immediately improved by AI use, there were no improvements in any of the functional performance variables measured when compared to the CN or sham conditions. These findings support previous studies which have attempted to investigate the effects of AIs on functional performance such as maximal strength and power output. Resistance trained individuals (> 2 y training experience) have previously demonstrated no improvement in isometric or dynamic contractions of submaximal or maximal intensity, following AI inhalation (14–17). These previous studies assessed strength using closed-chain exercises involving multiple muscle groups; in such cases participant familiarity with the exercise test is critical, even among well-trained individuals (36). The current study recruited healthy, recreationally active participants who were unfamiliar with resistance training techniques, as these individuals could be expected to receive a greater beneficial effect from increasing arousal (4). By adopting open-chain exercises, we were able to isolate single muscle groups to assess maximum isometric contraction with minimal participant learning effects (37) and greater sensitivity. Additionally, internal joint angles of 60° (KE) and 90° (HG) were selected to optimise isometric force (27, 28). Nonetheless, MVC, expressed relative to CN, did not differ between AI and sham conditions (KE: [-1.36, 7.04], HG: [-6.22, 4.87]).

Similar to previous research, we recorded no improvement in absolute maximal force production with AIs. However, aspects of force generation, such as RFD and EMD have previously been shown to improve following AI use (14), as well as with other stimulants, such as caffeine, both in the presence (38) and absence (39) of enhanced performance. Interestingly, Bartolomei et al. (2018) reported an association between RFD and CMJ performance, despite no effect of AI use on CMJ peak power (14). However, in contrast to the previous study we observed no effect of AIs on RFD, and further, no effect on EMD or RT. Similar RFD results have been reported previously elsewhere, as Perry et al. (2016) also reported no difference following AI use compared to a control (15). The inconsistency in RFD outcomes with AI use could stem from the type of exercise used to assess RFD – while both Perry et al. (2016) and Bartolomei et al. (2018) utilised maximal isometric mid-thigh pull, participants in the latter study sustained 6 s contractions (compared to 2 s contractions in the former) (14, 15). While our present study adopted 5 s contractions, these involved fewer muscle groups, and were separated by 5 min rest periods (compared to 3 min (14)). As such, and similar to during the study by Perry et al. (2016), participants in the present study may have been less subject to fatigue than were those involved in the study by Bartolomei et al. (2018). As previous findings have shown AIs to improve peak and mean power in fatigued individuals (13), so too it is possible that improvements in RFD may also be observable only in individuals exhibiting greater levels of fatigue. It is well established that RFD is impaired by fatigue, indeed such impairment is more pronounced than is impairment in strength (40). Fatigue-induced decline in RFD presents a greater potential for a stimulant-derived increase, therefore future research should explore the relative impact of AI use on aspects of force production, including RFD, in the presence of fatigue-induced decrements.

353 Contrary to our hypothesis, we observed no effect of AI use on peak power. Previously, peak
354 and mean anaerobic power have been seen to increase post-AI use (13). In that study
355 participants completed a 30 s Wingate anaerobic cycling test - given the large muscle mass
356 used in such a test, and the benefits of increased blood flow to working muscles in the latter
357 half of the test (41), elevated mean power could be explained by an AI-induced sympathetic
358 response. However, peak power would not be enhanced by such an increase in oxidative
359 metabolism. Unlike the present study, where participants were well rested prior to performing
360 CMJ, participants in the aforementioned study by Secrest et al. (2015) were required to reach
361 fatigue before the effects of AI use on anaerobic power was assessed (13). Similar to RFD; AI
362 use may only produce substantive effects on power performance when peak power has
363 previously been depleted by individuals reaching a state of fatigue, such as may be experienced
364 in the latter stages of sporting competition. Indeed, based on the results of their survey of
365 powerlifters, Pritchard et al. (2014) reported that use of AIs is most common near the end of
366 competition (12). Although existing evidence permits us to speculate that AI use may be able
367 to offset some of the decrement in RFD and peak power that is experienced alongside fatigue,
368 our findings demonstrate that in a well-rested state, there is no performance benefit to AI use,
369 despite elevated arousal and an associated perception of performance enhancement.

370

371 There are limitations which must be considered when interpreting our findings. Despite AIs
372 being held at a distance of 150 mm (in accordance with manufacture suggestions), it is possible
373 that participants did not inhale a consistent volume throughout the trial, however this closely
374 reflects real world practice. To the best of our knowledge, no research provides data regarding
375 the volume of substance inhaled from set distances. Whilst one similar study held AI 10cm
376 below the nose (14), other studies in this area did not identify a distance. As inhalation volumes
377 for each participant and test cannot be controlled for, we incorporated a distance of 15cm and

asked participants to sharply inhale when instructed to aid with intra- and inter-visit consistency. In addition, rapid rating of alertness was required to ensure that the observations represented participant alertness immediately after AI use, precluding the use of an extensive questionnaire, as such alertness was reported using VAS. VASs have been shown to be an effective method of collecting data efficiently, however, the limited time available to record alertness prevented us from collecting a rating from participants prior to every repetition (i.e., following every inhalation). A pragmatic decision was taken to record alertness during one repetition (from three) of each assessment, randomly selected. Furthermore, whilst we attempted to reduce potential learning effects by familiarising participants during the control visit and randomising the order of the experimental and sham conditions, it is possible that randomising the order of the strength and power performances during each visit may have reduced this risk further. Lastly, it much be acknowledged that EMD displayed poor reliability within this study. Therefore, the EMD results within this study should be interpreted with caution.

In conclusion, we have demonstrated for the first time that ammonia inhalant use elevates levels of alertness and perception of overall performance in active, non-resistance trained males. However, peak force and rate of force development, reaction time, and peak power all remain unaffected by ammonia inhalant use. Ammonia inhalants remain a popular sporting supplement, although perceived benefits may outweigh any true ergogenic effect; further research is required to explore the potential of acute ammonia inhalation to counter the performance-damaging effects of fatigue.

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Tables

Table 1. Mean (SD) rate of force development over 0-20, 0-50, 0-100 and 0-200 ms from onset of force production, electromechanical delay (EMD), and reaction time (RT), within the control condition (no inhalant) and post-inhalation within the experimental (ammonia) and sham conditions (water).

		Control	Ammonia	Sham
HG RFD ($N \cdot s^{-1}$)	0-20 ms	232.79 (260.23)	386.86 (434.09)	396.97 (395.75)
	0-50 ms	475.23 (445.69) *	683.29 (545.07) *	806.15 (638.21) *
	0-100 ms	958.43 (596.87) **†	1195.76 (639.31) **†	1445.52 (893.24) **†
	0-200 ms	1315.20 (464.45) **†	1442.78 (612.09) **†	1398.89 (481.44) **†
HG	EMD (ms)	55.14 (36.56)	58.54 (17.27)	49.57 (21.70)
	RT (ms)	339.86 (134.83)	297.36 (116.36)	280.86 (60.41)
KE RFD ($N \cdot s^{-1}$)	0-20 ms	294.10 (333.12)	537.14 (647.69)	312.91 (277.81)
	0-50 ms	570.85 (510.52) *	730.35 (612.35) *	579.35(453.77) *

	0-100 ms	676.03 (461.36) **	813.37 (487.33) **	652.94 (414.91) **
	0-200 ms	556.96 (298.20) ¶	452.77 (200.95) ¶	604.34 (281.76) ¶
KE	EMD (ms)	84.89 (30.88)	88.75 (46.45)	87.64 (27.22)
	RT (ms)	380.79 (129.58)	402.54 (150.68)	357.18 (84.99)

***HG = handgrip; KE = knee extension*.**

Significant main effect for time during rate of force development is represented by the following symbols: * = P < 0.05 from 20ms; ** = P < 0.001 from 20ms; † = P < 0.001 from 50ms; ¶ = P < 0.05 from 100ms.

Figures

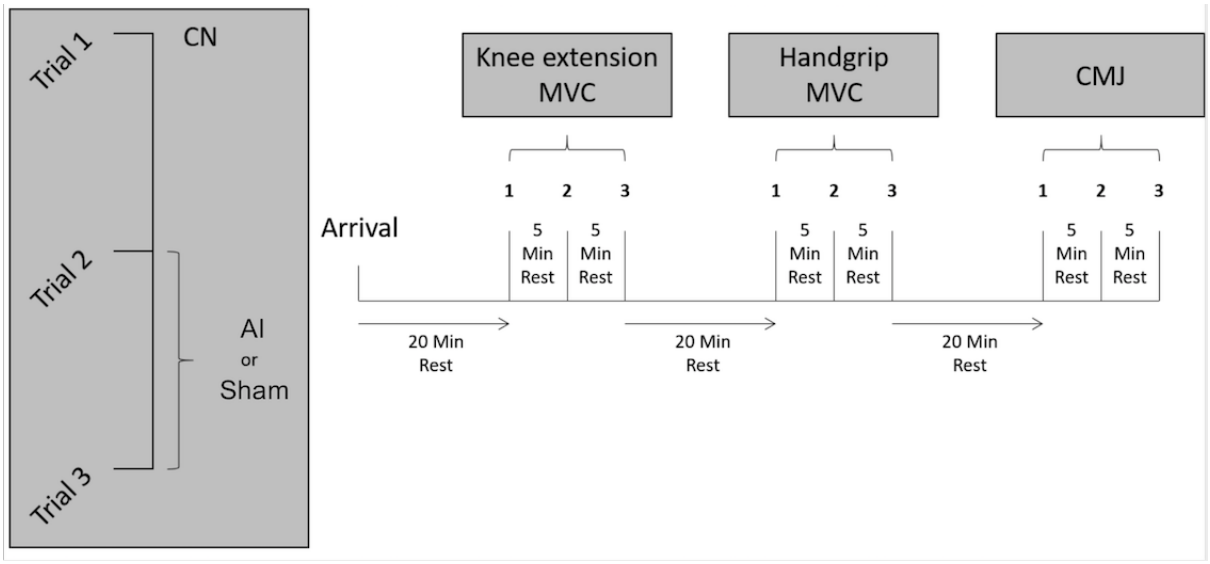
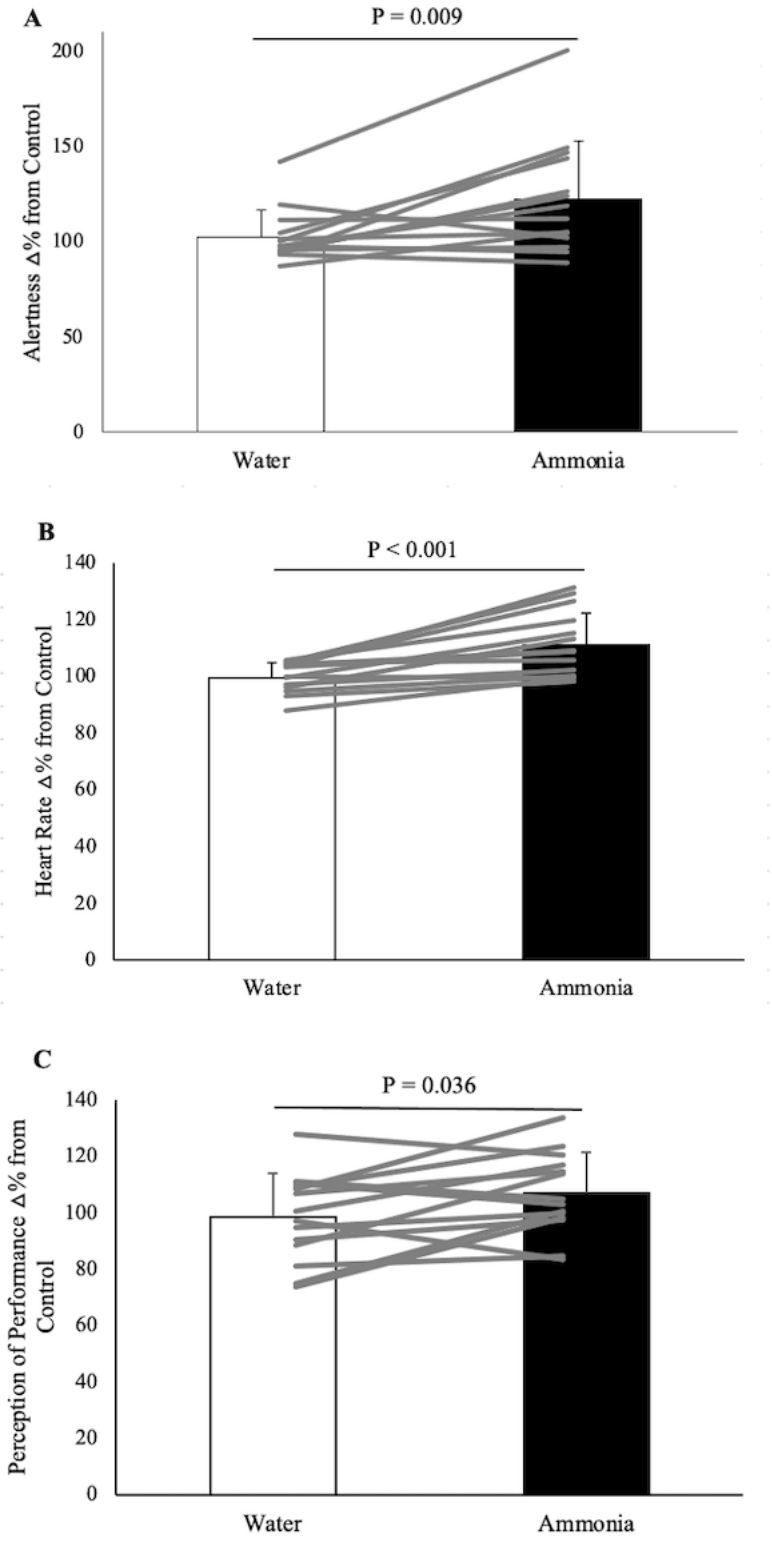


Figure 1. Timeline of measures during the 3 laboratory visits.
CN = control, AI = ammonia inhalant.
MVC = maximal isometric voluntary contraction, CMJ = countermovement jump.
Participants inhaled AI or water prior to each contraction and CMJ during trials 2 and 3. Heart rate and alertness were measured immediately post-inhalation prior to repetition 1, 2 or 3 (assigned at random) of knee extension MVC, handgrip MVC and CMJ.



552
553 Figure 2. Mean (SD) of heart rate (A), alertness (B), and perceived performance (C) presented
554 as the percentage difference from the control during the experimental (ammonia) and sham
555 (water) conditions.
556 *VAS = visual analogue scale*.
557

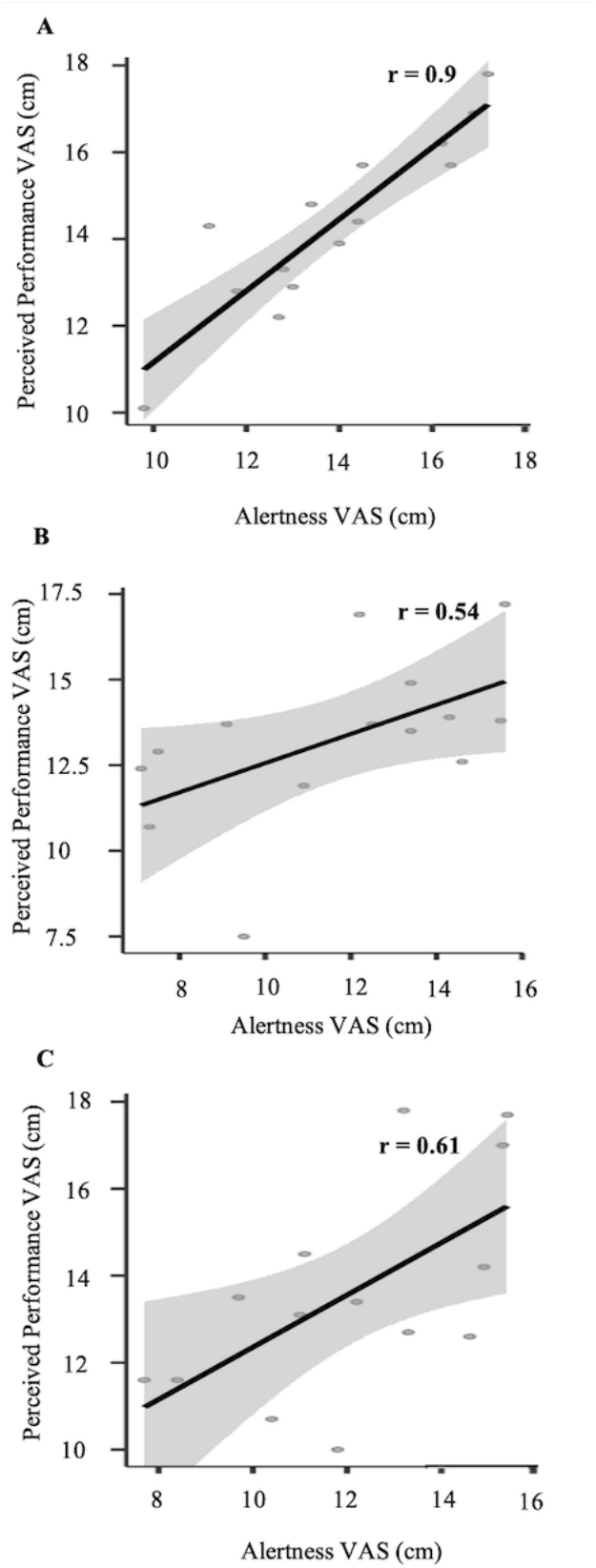


Figure 3. Relationship between alertness and perceived performance including 95% confidence intervals: (A) post-inhalation of ammonia (AI), (B) post-inhalation of water, (C) during control condition (no inhalant).

563 *VAS = visual analogue scale.*
564

565