SHORT COMMUNICATION

# Habituation of the Cold Shock Response May Include a Significant Perceptual Component

Martin J. Barwood, Jo Corbett,
and Christopher R. D. Wagstaff

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**Introduction:** Accidental immersion in cold water is a risk factor for many occupations. Habituation to cold-water immersion (CWI) is one practical means of reducing the cold shock response (CSR) on immersion. We investigated whether repeated thermoneutral water immersion (TWI) induced a perceptual habituation (i.e., could lessen perceived threat and anxiety) and consequently reduce the CSR on subsequent CWI. **Methods:** There were 12 subjects who completed seven 7-min head-out immersions. Immersions one and seven were CWIs [15.0 (0.1)°C], and immersions two to six were TWI [34.9 (0.10)°C]. Anxiety (20-cm visual analogue scale) and the cardiorespiratory responses [heart rate ($f_c$), respiratory frequency ($f_R$), tidal volume ($V_T$), and minute ventilation ($\dot{V}_E$)] to immersion were measured throughout. Data were compared within subject between conditions using ANOVA to an alpha level of 0.05. **Results:** Acute anxiety was significantly reduced after repeated exposure to the immersion scenario (i.e., TWI): CWI-1: 6.3 (4.4) cm; and CWI-2: 4.5 (4.0) cm [condition mean (SD)]. These differences did not influence the peak in the CSR. The $f_c$, $f_R$, and $\dot{V}_E$ responses were similar between CWI-1 and CWI-2. $V_T$ response was significantly lower in CWI-2; mean (SD) across the immersion: CWI-1 1.27 (0.17) vs. CWI-2 1.11 (0.2) L. **Discussion:** Repeated TWI lessened the anxiety associated with CWI (perceptual habituation). This had a negligible effect on the primary components of the CSR, but did lower $V_T$, which may reduce the volume of any aspirated water in an emergency situation. Reducing the threat appraisal of an environmental stressor may be a useful biproduct of survival training, thereby minimizing psychophysiological strain.

**Keywords:** helicopter underwater escape, cold shock response, habituation, anxiety.

ACCIDENTAL COLD-water immersion (CWI) is a risk factor for aviators and aircrew, for persons who work on or around cold water, and the general public. The responses evoked by whole body CWI are life threatening and are described collectively as the "cold shock response" (CSR). The CSR is triggered by a rapid change in skin temperature and is characterized by an initial inspiratory gasp followed by uncontrollable hyperventilation and tachycardia, which in combination impose a significant cardiorespiratory strain. In otherwise healthy individuals the loss of respiratory control is of primary concern in the early minutes of immersion, increasing the risk of aspirating water and drowning (10). Using maximal breath-hold time as an index of respiratory control, only 34% of subjects completing offshore survival training could produce a breath-hold in cool water that was sufficient to enable them to egress a ditched and inverted helicopter in ideal conditions (5). It is the shortfall between the maximum breath-hold time of individuals in cold water and the time required to make an underwater escape from a helicopter [28-92 s (5)] which provides the rationale for the use of survival aids that reduce the CSR or protect the airway until CSR subsides [i.e., immersion dry suits and Emergency Underwater Breathing Systems (12)]. However, survival aids are not always available if immersion is sudden and in these circumstances it is prudent to identify other means of reducing the CSR.

The CSR can be influenced by positive and negative psychological components (1,2). Indeed, subjects who received a psychological skills training (PST) intervention improved (positive/beneficial effect) their maximal breath-hold time on immersion by 80% (2). In contrast, in a separate recent study, components of the CSR (i.e., minute ventilation and heart rate) were increased (negative effect), which may increase the risk of drowning, when subjects were immersed in a hyper-anxious state (1). The psychological component of the CSR has also been studied after repeated CWIs, which induces a habituation (3). When PST was combined with habituation, subjects increased their maximal breath hold time by 26.86 (24.70) s. This beneficial effect equated to a 120% improvement on their maximal breath-hold time in a control immersion. Importantly, the extent of the improvement did not exceed that of an "habituation alone" group who simply underwent repeated CWI and did not receive any psychological support (3). This raises the possibility that repeated experience of the immersion scenario per se, as was the case with the "habituation alone" group, in itself confers a psychological benefit similar to that of PST. Theoretically subjects could evaluate the immersion scenario as being increasingly threatening or nonthreatening depending on their appraisal of the psychological demands of the situation. It follows that those studies that have examined the effect of anxiety on the CSR, before and after habituation,

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From the University of Portsmouth, Department of Sport and Exercise Science, Portsmouth, U.K.

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Address correspondence and reprint requests to: Dr. Martin Barwood, Department of Sport and Exercise Science, University of Portsmouth, Spinnaker Building, Cambridge Road, Portsmouth PO1 2ER, UK; martin.barwood@port.ac.uk.

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indicate a change in appraisal (1,8) of the threat posed by imminent immersion.

In these studies, subjects were deceived about the expected water temperature as being 5°C colder than experienced previously; in reality it was unchanged (1). The consequence of this reappraisal was the negative emotional state of anxiety and a magnified CSR or a reversal of the previously habituated component of the CSR, both of which could be negative in a real-life scenario. However, it has yet to be established whether habituation of this threat-perception carries a physiological benefit for the CSR. Accordingly, we hypothesized that the CSR would be reduced after repeated exposure to the immersion scenario in the absence of a cold-water thermal stimulus which is known to, in part, produce an habituation. In short, we sought to separate the thermal and perceptual implications of habituation; anxiety rating was used as a surrogate of reduced threat perception and a change in appraisal of threat.

## METHODS

### *Subjects*

The study protocol was approved by the Biosciences Research Ethics Committee and the subjects gave their written informed consent. There were 12 healthy, nonsmoking subjects (8 men, 4 women) who volunteered for the experiment [mean (SD); age 20 (1) yr; height 1.72 (0.10) m; mass 70.48 (14.95) kg]. The subjects were nonsmokers, were not cold-water habituated, and were naïve to the aims of the experiment. They were asked to abstain pretest from alcohol and caffeine consumption for 24 h.

### *Experimental Design*

The study used a within-subject repeated measures design. The subjects visited the laboratory on seven separate occasions to complete seven whole-body water immersions; two CWI [immersions 1 and 7; water temperature ($T_{water}$ 15°C)] and five habituation immersions into thermoneutral (35°C) water. Immersion (IMM) 1 and 7 took place at the same time of day to minimize circadian variation. IMM 2 to 6 were thermoneutral water immersions (TWI) and were performed in order to habituate the perceptual threat component associated with the immersion scenario; these were completed on separate days between 09:00 and 17:00.

### *Procedure*

Following arrival at the Extreme Environments Laboratory, each subject's height (m) and mass (kg) were recorded using a stadiometer (Bodycare Stadiometer, Leicester, UK) and calibrated weighing scales (OHAUS digital weighing scales, Parsippany, NJ). Each subject changed into a swimming costume; the same swimming costume was worn by the subject on each occasion. Subjects were then instrumented with a 3-lead ECG (HME Lifepulse, Derbyshire, UK) and entered an ambient temperature ($T_a$) controlled laboratory. They sat on an immersion chair attached to an electronic winch (CPM, F1-8; 2-8; 5-4, Yale, Shropshire, UK) with a seat belt fastened around their waist to counteract buoyancy. The subject inserted a two-way mouthpiece (Harvard Instruments, Harvard, MA) and attached a noseclip. The mouthpiece was connected to a spirometer (spirometric transducer module, KL Eng. Co, Northridge, CA) by respiratory tubing in order to measure the respiratory responses to immersion. The subject was winched above the immersion tank to rest for 1 min. They provided their acute anxiety rating 30 s into the 1-min rest period on a visual analogue scale; they were familiarized with the scale in advance of the study. Toward the end of the 1-min period a 10-s verbal countdown preceded the subject being lowered at a reproducible rate ($8 \text{ m} \cdot \text{min}^{-1}$) until immersed to the clavicle; immersion depth was standardized within subject on each occasion. After 1, 3, 5, and 7 min of immersion they again reported their anxiety rating, following which they were winched from the immersion tank.

### *Measurements*

Water temperature ($T_w$) and $T_a$ were measured and recorded using a calibrated thermistor [Grant Instruments (Cambridge) LtD, Shepreth, UK] secured to the wall of the immersion tank and a wet bulb globe thermometer station, respectively, both attached to a data logger [1000 series, Squirrel Data Logger, Grant Instruments (Cambridge)]. Average $T_w$ was closely matched within subject (± 0.1°C) between CWIs; $T_w$ CWI-1: 15.0 (0.1)°C; and CWI-2: 15.0 (0.2)°C. The average $T_a$ during the CWIs was: CWI-1: 22.6 (2.1)°C; and CWI-2: 21.0 (2.10)°C. $T_w$ and $T_a$ during TWI averaged 34.9 (0.1)°C and 24.7 (0.2)°C, respectively, across the five immersions.

The ECG and spirometer were interfaced with a digital data acquisition system (16SP PowerLab, Castle Hill, Australia) which captured data continuously throughout the rest and immersion periods. Chart analysis software (Chart version 6, AD Instruments, Axminster, Devon, UK) was used to automatically identify R-waves from the ECG and calculate cardiac frequency ($f_c$); movement artifacts were visually identified and excluded from analysis. The spirometer was calibrated using a syringe of known volume (3-L syringe, Harvard Instruments, Harvard, MA). Respiratory frequency ($f_R$) was recorded by Chart analysis software using auto-recognition of the peak after inspiration. The peak value after the onset of inspiration was recorded as tidal volume ($V_T$) and multiplied by the calculated $f_R$ to generate minute ventilation ($\dot{V}_E$). The state anxiety response to immersion was quantified using a 20-cm visual analogue scale with descriptive phrases ranging from 0 cm (not at all anxious) to 20 cm (extremely anxious).

### *Data Analyses*

The normality of the data was checked. With the exception of the state-anxiety data, the analyses were focused on the CWI responses. The magnitude of the CSR was examined by visually identifying the absolute peak value (i.e., the highest 'true' value generated between two consecutive breaths) in $f_c$ and $f_R$ that occurred immediately prior to immersion and on immersion. The duration of the cold shock was examined by generating 1-min averages for the pre-immersion phase and for each 1-min period of the 7-min immersion. Univariate analyses were checked for sphericity using Mauchley's test and, where nonspherical data sets were evident, a Greenhouse-Geisser adjustment was applied. The direction of statistically significant effects was determined using a post hoc pair-wise comparisons procedure. For all statistical tests $\alpha$ level was set at 0.05. Data are presented as mean (SD). All statistical tests were conducted using SPSS version 18 (Chicago, IL).

The $T_w$ and $T_a$ during the CWI were compared using an independent samples *t*-test. The anxiety scores pre-immersion and from minutes 1, 3, 5, and 7 were compared across all immersions using factorial ANOVA [condition (7) × time (5)]. The peak in CSR ($f_c$ and $f_R$) in each CWI were examined using a repeated measures ANOVA [condition (2) × time (2)]. The duration of CSR was examined using the 1-min average data for $f_c$, $f_R$, $V_T$, and $\dot{V}_E$ pre- and on immersion using a separate repeated measures factorial ANOVA [condition (2) × time (8)].

## RESULTS

There was no significant difference in the $T_w$ ($t = 1.958$, $P = 0.537$) or $T_a$ ($t = 0.963$, $P = 0.903$) between CWI-1 and CWI-2. Subject's state anxiety peaked prior to immersion and gradually declined from minute 1 to minute 7 of the immersion irrespective of water temperature [significant main effects for time; $F(4,44) = 11.695$, $P = 0.001$]. The anxiety experienced prior to and during CWI was significantly greater than that evident during TWI [significant main effects for condition; $F(6,66) = 16.247$, $P = 0.001$]. Anxiety associated with TWI gradually declined from TWI 1 to TWI 3, following which there were no differences in the anxiety reported, indicating a plateau in the response and a perceptual habituation. There were significant differences in the anxiety experienced during CWI-2 being lower overall than CWI-1 ($P = 0.013$). Time point specific differences were also evident at minutes 1 ($P = 0.025$), 3 ($P = 0.042$), 5 ($P = 0.001$), and neared being different after 7 min of immersion ($P = 0.052$); pre-immersion the anxiety ratings were similar ($P = 0.296$). Interaction effects were also evident [$F(24,264) = 4.574$, $P = 0.008$]. State anxiety responses are summarized in **Fig. 1**.

The peak in the CSR during CWI occurred on immersion in both $f_c$ [$F(1,11) = 62.117$, $P = 0.001$] and $f_R$ [$F(1,11) = 48.505$, $P = 0.001$]. The significant differences in anxiety did not influence the peak in the CSR [no main effect for condition: $f_c$, $F(1,11) = 0.001$, $P = 0.986$; $f_R$, $F(1,11) = 0.471$, $P = 0.507$] in anticipation of immersion [peak $f_c$ CWI-1: 105 (17) vs. CWI-2 105 (19) bpm; and $f_R$ 32 (8) vs. 0.31 (9) breaths $\cdot$ $\text{min}^{-1}$] or on immersion [peak $f_c$ was CWI-1: 126 (19) vs. CWI-2 127 (19) bpm; and $f_R$ 84 (28) vs. 0.80 (31) breaths $\cdot$ $\text{min}^{-1}$]. There were no interaction effects [$f_c$, $F(1,11) = 0.034$, $P = 0.857$; $f_R$, $F(1,11) = 0.186$, $P = 0.657$].

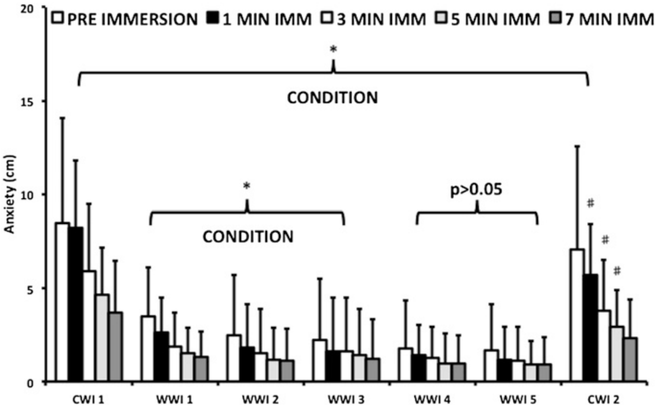

**Fig. 1.** Mean (SD) acute anxiety response to CWI-1 and 2 and TWI 1 to 5; *indicates significant difference between conditions; #indicates significant difference between CWI-1 and CWI-2 ($N = 12$).

Consistent with the state anxiety responses and peak CSR data, the 1-min averaged CSR data increased from pre- to on immersion and gradually declined as the immersion ensued (significant main effects for $f_c$, $f_R$, $\dot{V}_E$, and $V_T$; $P < 0.05$). Each minute across the immersion the $f_c$, $f_R$, and $\dot{V}_E$ responses were similar between CWI-1 and CWI-2 (no significant main condition effect for these variables; $P > 0.05$; **Table I**). Similarly, $f_c$, $f_R$, and $\dot{V}_E$ also showed no interaction effect ($P > 0.05$), but the $V_T$ response decreased at a significantly faster rate in CWI-2 [main interaction effect $F(7,77) = 2.970$, $P = 0.041$]. Timepoint specific differences were evident after 2 ($P = 0.025$) and 6 ($P = 0.046$) min of immersion, neared being different at 4 min ($P = 0.069$) of immersion, and had a tendency to be lower throughout (**Fig. 2**) [mean (SD) across the immersion: CWI-1: 1.27 (0.17) vs. CWI-2: 1.11 (0.2) L].

## DISCUSSION

This study examined the possibility that the CSR could be reduced by habituation of the perceptual component of the CSR alone in the absence of any repeated cold-water stimulation. The significant difference in anxiety data during CWI-2 (Fig. 1), as a consequence of repeated TWI, are consistent with the idea that subjects began to evaluate the immersion scenario per se as less threatening. This perceptual change did not induce any significant alteration in the CSR peak or in the majority of CSR variables. However, the ventilatory component of the CSR ($V_T$) was significantly lower in CWI-2 than CWI-1, which suggests the lower anxiety ratings in CWI-2, at least in part, culminated in an altered physiological response. This difference was not of a sufficiently consistent magnitude, when coupled with $f_R$ data, to equate to an increase in $\dot{V}_E$. Collectively, these data enable our hypothesis only to be partly accepted.

Previous literature suggests an increase in acute anxiety has the potential to magnify the $f_c$ and sustain the ventilatory components of the CSR, on average, even after habituation has taken place (1). However, it seems that an increase in state anxiety has a greater influence on components of the CSR, irrespective of habituation,

**TABLE I. MEAN (SD) OF 1-min AVERAGED $f_c$, $f_R$, AND $\dot{V}_E$ RESPONSES IN CWI-1 AND CWI-2 ($N = 12$).**

| | PRE | MIN 1 | MIN 2 | MIN 3 | MIN 4 | MIN 5 | MIN 6 | MIN 7 |
|---|---|---|---|---|---|---|---|---|
| CWI-1 $f_c$ (bpm) | 91 (17) | 95 (19) | 90 (19) | 87 (18) | 83 (16) | 81 (15) | 80 (13) | 78 (15) |
| CWI-2 $f_c$ (bpm) | 93 (21) | 91 (21) | 89 (19) | 85 (17) | 84 (16) | 82 (15) | 80 (15) | 81 (15) |
| CWI-1 $f_R$ (breaths · $\text{min}^{-1}$) | 19 (4) | 29 (7) | 21 (5) | 20 (5) | 19 (5) | 20 (5) | 21 (7) | 21 (6) |
| CWI-2 $f_R$ (breaths · $\text{min}^{-1}$) | 19 (4) | 27 (8) | 21 (8) | 20 (4) | 19 (4) | 19 (4) | 19 (5) | 19 (4) |
| CWI-1 $\dot{V}_E$ (L · $\text{min}^{-1}$) | 16.8 (4.3) | 41.1 (13.9) | 28.5 (12.2) | 24.0 (10.3) | 23.6 (12.4) | 23.50 (14.1) | 24.0 (14) | 23.2 (15.4) |
| CWI-2 $\dot{V}_E$ (L · $\text{min}^{-1}$) | 17.2 (3.2) | 40.6 (15.4) | 24.8 (9.5) | 21.1 (7.0) | 19.0 (7.2) | 18.1 (6.0) | 17.1 (6.1) | 17.8 (5.6) |

than the modest reduction seen with the habituation of the perceptual component of the response as demonstrated in the present study. The findings of the present study also contextualize the data of Barwood et al. (3), who suggested that CSR habituation may include a perceptual component. The lack of difference in maximal breath-hold time (a surrogate of respiratory control) between an 'habituation alone' and an habituation plus PST group observed in their study appeared to suggest the presence of an inherent perceptual and evaluative component to an habituation regimen.

Previous studies examining habituation of the cold pressor response during hand immersion have suggested that perceptual afferent information, in the form of an increase in anxiety, arriving for processing at the same time as thermal afferent information, has the potential to disrupt the central nervous system processing of the temperature related sensory information (7). We supported this idea in whole body immersion and suggested a role for the amygdala in appraising the emotional valence of the environmental stimuli prior to and on immersion (1), and the present and the previous studies now seem to highlight a need to confirm this idea. In support, it is known that the amygdala is involved in the central nervous system response to psychological stress and anxiety. It is also known that the amygdala projects to the dorsomedial hypothalamus, which provides at least one viable route by which the efferent response during CWI may be influenced (6). From a psychological perspective, current stress theory would suggest that repeated exposure to the immersion scenario would culminate in a change in the primary (i.e., importance, novelty) or secondary (i.e., coping resources) appraisal of the immersion scenario as threatening (8). The present study suggests that this, in part, has some physiological consequence.

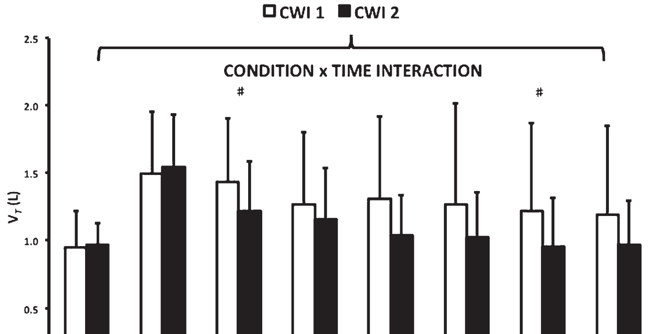

**Fig. 2.** Mean (SD) $V_T$ response to CWI-1 and 2; #indicates significant difference between CWI-1 and CWI-2 ($N = 12$).

This study is not without limitation. Indeed, the study lacks a distinct control group to directly test the hypothesis that the anxiety rating and, therefore, the threat perception, is not naturally lower on secondary CWI in contrast to an initial CWI. However, this idea can be roundly rejected based on evidence from other studies that have examined the anxiety response over consecutive immersions (1) or examined the extent of the CSR across two CWIs (11). Indeed, it seems that an increased or unchanged anxiety rating is more likely in consecutive immersions accompanied by an increased or matched CSR (2). Other prominent researchers in this area concur that, possibly due to recalibration of expectation after initial immersion (subjects often report greater perceived CSR than expected in the first CWI), the CSR on secondary immersion is more likely to be greater than lesser (Prof. M. J. Tipton; personal communication; May 2013). The reverse of that was evident in the present study. One such solution for this oversight would be to match subjects based on their initial CWI response and examine for any differences in the response with and without five TWI between two CWIs. However, it is difficult to determine exactly which CSR matching criteria should be used and, coupled with other literature evidence, this seems unnecessary. The findings are also limited to the specific cohort we tested; variations associated with selection bias, swimming capability, gender, ethnic group, and occupational background cannot be uncovered by the data produced by the present study.

The practical implications of our findings are important. Indeed, given that the aspiration of as little as 22 ml · $\text{kg}^{-1}$ of seawater can be fatal [1.65 L for an average 75-kg individual (9)], even the relatively small reduction in $V_T$ seen in the present study as a consequence of the perceptual habituation could be meaningful in reducing the potential volume of water aspirated on accidental CWI. It is also possible that perceptual habituation may be achieved through repeated exposure to other emergency test scenarios such as survival training. Indeed, helicopter underwater escape training (HUET) may represent a means of inducing threat reappraisal of a ditch scenario and, consequently, the associated anxiety in a real-life situation. Consistent with this idea, Tipton et al.

(11) reported lower heart rate responses to repeated HUET tests (a possible perceptual habituation) in the absence of any repeated CWI. Similarly, Brooks et al. (4) treated a subject who was excessively anxious and underperforming in HUET tests by repeatedly exposing him to parts of the test scenario and reducing the associated anxiety. With this treatment, the subject eventually passed the HUET test course. Based on evidence from the work of Tipton et al. (11) and Brooks et al. (4) and that reported in the present study, it may be that survival training carries a perceptual habituation and consequent reduction in the physiological response to a given test scenario that carries over to an emergency situation. However, we are not suggesting that thermoneutral immersion is entirely sufficient to enable habituation; it probably comprises only a small part of the response. Therefore, replicating the likely environmental conditions as closely as possible during survival training is most likely to confer a benefit in an emergency scenario.

In summary, our data show that repeated exposure to the immersion scenario, in the absence of repeated cold-water stimulation, reduces the ventilatory component of the CSR. Given that surviving an accidental immersion is decided by fine margins, this difference could be meaningful in a real-life scenario.

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*Authors and affiliation:* Martin J. Barwood, B.Sc., Ph.D., Jo Corbett, M.Sc., Ph.D., and Christopher R. D. Wagstaff, M.Sc., Ph.D., Department of Sport and Exercise Science, University of Portsmouth, Portsmouth, UK.

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