Post-exercise cold-water immersion activates acute PHF20 and p53 signalling in human skeletal muscle

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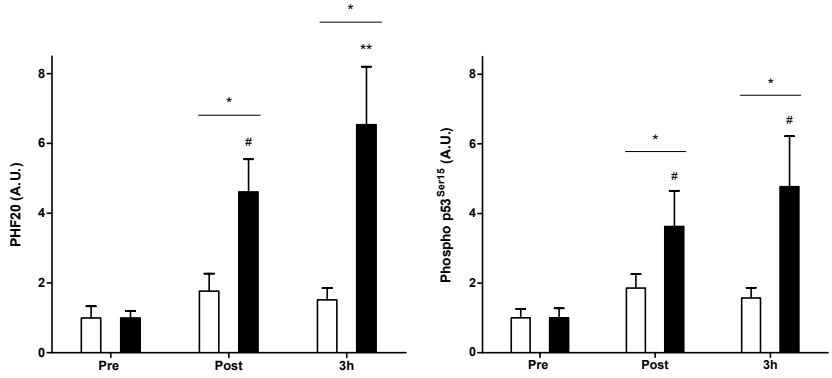
Despite its widespread use in post-exercise recovery (Broatch *et al.*, 2014), debate currently exists surrounding the merit of cold-water immersion (CWI) in athletic training regimes. Short-term improvements in recovery from exercise may be thwarted by unfavourable long-term skeletal muscle adaptations (Yamane *et al.*, 2006). The aim of this study was to investigate the underlying molecular mechanisms by which CWI may alter the signalling pathways associated with mitochondrial biogenesis following an acute bout of high-intensity interval exercise.

Nineteen males (mean ± SD; age 24 ± 6 y; $\dot{V}O_{2peak}$ 46.5 ± 8.1 mL•kg^{-1}•min^{-1}) performed an acute high-intensity interval training (HIT) bout, comprising 4 × 30s all-out efforts on a cycle ergometer, immediately followed by one of two 15 min recovery conditions: CWI (10.3 ± 0.2°C) or a passive control at ambient room temperature (CON; 23 ± 0.1°C). Muscle biopsies (*vastus lateralis*) were obtained pre-exercise, post-recovery and 3 h post-recovery to determine the acute molecular signalling response following HIT and CWI. Phosphorylation (p-) of p38 MAPK$^{Thr180/182}$ (3.0 ± 0.9 *vs* 2.4 ± 0.6), AMPK^{Thr172} (2.7 ± 0.8 *vs* 5.5 ± 1.6) and p53^{Ser15} (1.9 ± 0.4 *vs* 3.6 ± 1.0) increased immediately post-recovery ($P < 0.05$) in CON and CWI, respectively. p-p38 MAPK returned to basal levels by 3 hours post-recovery, whereas p-AMPK (2.3 ± 0.5 *vs* 6.5 ± 2.6) and p-p53 (1.6 ± 0.3 *vs* 4.8 ± 1.5) remained significantly elevated for both conditions ($P < 0.05$). When compared with CON, CWI resulted in larger increases in p-p53 (ES = 0.92, $p = 0.058$) and the content of its upstream regulator PHF20 ($P < 0.05$) immediately post-recovery and 3 h post-recovery (Figure).



*Total PHF20 protein and phosphorylation of p53^{Ser15} immediately pre-exercise (Pre), post-recovery (Post), and 3 h post recovery (3h) for CON (open bars) and CWI (closed bars) conditios. *Significant difference from pre-exercise (P < 0.05). **Significant difference from CON. #Large effect (ES > 0.8) from CON. Data are presented as mean ± S.E.M.*

We provide novel data demonstrating that post-exercise CWI alters acute molecular signalling pathways associated with mitochondrial biogenesis. Recently implicated as an important regulator of mitochondrial function (Saleem *et al.*, 2011), p53 activation following CWI may serve as a novel and potent stimulus by which to enhance contraction-induced mitochondrial biogenesis. Our findings are consistent with reports of post-exercise CWI increasing the expression of peroxisome proliferator-activated receptor gamma (PPARγ) coactivator 1-alpha (PGC-1α) (Ihsan *et al.*, 2014). The mechanisms by which increases in PHF20 and p53occur following CWI are currently unknown, but may be related to the cellular stress imposed by a hypothermic shock and subsequent rewarming.

Broatch JR, Petersen A, Bishop DJ. (2014) *Medicine & Science in Sports & Exercise*, In press.

Ihsan M, Watson G, Choo HC, Lewandowski P, Papazzo A, Cameron-Smith D. Abbiss CR. (2014) *Medicine & Science in Sports & Exercise*, **46**: 1900-1907.

Saleem A, Carter HN, Iqbal S. Hood DA. (2011) *Exercise and Sport Sciences Reviews*, **39**: 199-205.

Yamane M, Teruya H, Nakano M, Ogai R, Ohnishi N. Kosaka M. (2006) *European Journal of Applied Physiology*, **96**: 572-580.

This study was partly funded by an Exercise & Sports Science Australia (ESSA) Applied Sports Science Research Grant (2011).