Effects of antioxidant supplementation and exercise training on skeletal muscle antioxidant enzymes and mitochondrial biogenesis

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An acute endurance exercise bout increases free radical production in skeletal muscle. This 'hormesis' stimulates positive adaptations in skeletal muscle, leading to improvements in endurance performance. Nutritional supplement use is popular among athletes, yet paradoxically, antioxidant supplementation may not be beneficial. Vitamin E and α-lipoic acid are lipid and water soluble antioxidants, respectively and it is currently unknown whether these antioxidants inhibit muscular adaptations to endurance training. We investigated the role of redox regulation on changes in skeletal muscle following endurance exercise training.

Male Wistar rats (n=48) were divided into sedentary control (SC), sedentary + antioxidant (SA), exercise (E) and exercise + antioxidant (EA) groups. The antioxidant groups were supplemented with Vitamin E (1000 IU/kg diet) and α-lipoic acid (1.6 g/kg diet) for 14 wk. Exercising animals were treadmill trained (90 min/d, 4 d/wk at 70% VO_2max) during this time. Red gastrocnemius and vastus muscles were excised under general anaesthesia 48 h after the final training bout. Antioxidant enzymes (xanthine oxidase, XO; manganese superoxide dismutase, Mn-SOD; glutathione peroxidase, GPX), antioxidant enzyme gene expression (Mn-SOD and GPX-1), markers of mitochondrial biogenesis (peroxisome proliferator-activated receptor gamma, coactivator alpha, PGC-1α; mitochondrial transcription factor A, mtTFA; citrate synthase) and PGC-1α protein abundance was analysed. All data were tested for normality and a two way ANOVA was performed, with a Tukey *post hoc* analysis. Significance was assumed when $p < 0.05$.

Antioxidant supplementation significantly reduced skeletal muscle PGC-1α mRNA (main effect for antioxidant, $p < 0.05$), whereas exercise training significantly increased PGC-1α mRNA (main effect for exercise, $p < 0.05$). However, there was no significant effect of antioxidant treatment on the exercise-induced increase in PGC-1α mRNA. Furthermore, PGC-1α protein abundance was significantly increased after training (main effect for exercise, $p < 0.01$), although there was no significant effect of antioxidant supplementation. Citrate synthase, mtTFA, GPX and Mn-SOD gene expression was similar between the groups. There were significant ($p < 0.05$) antioxidant × exercise interaction effects for all antioxidant enzymes. When compared to the sedentary group, exercise training suppressed XO activity (E *vs.* SC; -0.3×; $p < 0.01$), whilst antioxidant supplementation reduced XO activity in sedentary group (SA *vs.* SC; -0.5×; $p < 0.001$). GPX activity increased after training in supplemented animals (EA *vs.* SA; 1.4×; $p < 0.01$) but was reduced in the sedentary, supplemented group (SA *vs.* SC; -0.3×; $p < 0.01$). Lastly, exercise training reduced Mn-SOD and total SOD activities (E *vs.* SC; -0.5×; -0.4×; $p < 0.01$, respectively). Antioxidant supplementation reduced Mn-SOD and total SOD activities only in sedentary muscles (SA *vs.* SC; -0.4×; -0.4×; $p < 0.001$, respectively).

In summary, antioxidant supplementation suppressed gene transcription of the mitochondrial biogenesis marker PGC-1α, but did not alter PGC-1α protein abundance. There was no effect of antioxidant supplementation on the exercise-induced increase in PGC-1α mRNA and protein levels. Antioxidant supplementation decreased the activities of antioxidant enzymes, with a variable effect of endurance training.