

## ACUTE COCOA FLAVANOL INTAKE AFFECTS ANTIOXIDATIVE CAPACITY AND MEDIATES THE NO- PATHWAY IN WELL TRAINED ATHLETES: IMPLICATIONS FOR EXERCISE PERFORMANCE.

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*Introduction*: Evidence exists that cocoa flavanol (CF) can increase nitric oxide synthesis. This promotes vasodilation and several physiological processes, such as mitochondrial respiration and biogenesis, glucose uptake and SR-Ca handling, that may support exercise performance<sup>1</sup>. Moreover, CF intake is also known for its antioxidative capacities (in vitro and in vivo), resulting in reduced levels of oxidative stress<sup>2</sup>. During exhaustive exercise, free radical production increases dramatically, resulting in an altered redox state in the muscle, which possibly inhibits muscle contraction<sup>3</sup>. Acute intake of antioxidants may help neutralize free radicals and therefore directly prevent a decline in exercise performance<sup>4</sup>. Moreover, increased antioxidative capacity may repress the formation of peroxinitrite, which may elevate NO availability<sup>5</sup>. The aim of this study was to examine the effect of CF intake on 1) NO production in response to exhaustive exercise, 2) exercise-induced changes in antioxidative capacity and oxidative stress and 3) exercise performance in well-trained athletes.

*Materials and methods*: Twelve well-trained men(PL3<sup>6</sup>) (mean  $\pm$  SD age, height, mass, VO<sub>2max</sub>: 30  $\pm$  3 years, 177.9  $\pm$  8.8 cm, 72.8  $\pm$  7.8 kg , 63.0  $\pm$  3.5 ml/kg/min) participated in this randomized, double-blind, cross-over study. Participants performed two 30-min time trials (TT) 1.5 and 3 hours after CF intake (900 mg) or placebo intake (PL 13 mg CF), interposed by passive rest. Lactate, glucose, heartrate, rate of perceived exertion (RPE) and power output were measured during the TTs. Blood samples were drawn at baseline, before and immediately after the two TTs and analysed for trolox equivalent antioxidative capacity (TEAC), Uric acid (UA), malonaldehyde (MDA), arginine and citrulline. TT performance was compared between PL and CF by paired t-test. Pacing strategy during TT and blood markers were analysed by repeated measured ANOVA.

*Results*: Time to complete TT1 tended to be lower after CF intake compared to PL intake  $(29'13'' \pm 1'19'' \text{ CF vs. } 29'47'' \pm 1'58'' \text{ PL}, p=.09)$ . After 25 minutes, a significant higher power output was observed in CF compared to PL. No differences in TT2 performance was observed between CF and PL. Glucose, lactate, heartrate and RPE increased significantly during both TTs, but were unaffected by CF intake. TEAC and UA significantly increased after both TTs. CF intake increased TEAC and UA, compared to PL intake (Figure 1A). MDA was increased by

exercise, but remained unaffected by CF intake. Arginine was significantly lowered by exercise. Before TT1, CF intake tended to increase arginine, compared to PL intake. After TT1, arginine levels did not differ between CF and PL. Citrulline significantly increased by exercise, but was not altered by CF intake (Figure 1B).

*Conclusion*: Acute CF intake tended to enhance performance on a TT 1.5 hours after intake. Increased power output was detected at the end of the TT. Exercise acutely increased antioxidative capacity, lipid peroxidation (marker of oxidative stress) and NO production (increased citrulline and decreased arginine). CF intake increased antioxidative capacity, but did not alter lipid peroxidation. CF intake tended to increased arginine levels, known to augment NO production and possibly explaining the improved exercise performance.



Figure 1. Relative changes in trolox equivalent antioxidative capacity (TEAC) (A) and arginine (B), compared to baseline (BL) in response to 2 time trials (TT1, TT2), 1.5 and 3 hours after cocoa flavanol (CF; dashed lines) or placebo (PL; full lines) intake. \*: significant effect of time (exercise) in 2x5 repeated measures ANOVA (different compared to pre-TT(1 or 2). \$: significant effect of drink (CF) in 2x5 repeated measures ANOVA (CF different from PL), p<0.05.

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