Acute *vs.* Chronic Citrulline Malate Supplementation on Muscle Fatigue

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ABSTRACT

Citrulline malate has been proposed to aid in reducing fatigue by increasing blood flow through promoting an increase in the nitric oxide synthase pathway along with the ability to remove ammonia and lactate accumulations. Results on the effectiveness of an acute supplementation are mixed, but it is proposed that regular consumption may help to attenuate the onset of fatigue during exercise. **PURPOSE**: To investigate the effects of acute and chronic citrulline malate supplementation on fatigue rate of the quadriceps. **METHODS:** Recreationally trained males (n=18, 24±5 yr, 83±14 kg, 174±6 cm) participated in seven testing sessions. The familiarization session consisted of participants performing a graded exercise test to determine max power output. In a randomized, counterbalanced order, participants consumed a placebo (PL) and citrulline malate (CM) treatment for two separate dosing periods. For each dosing period, participants reported on three separate days with seven days between each visit. The first experimental testing session for each dosing period was considered the baseline day (BL), the second session the acute day (D1), and the third session the chronic day (D2). For chronic supplementation, all participants consumed each treatment for seven consecutive days. The exercise protocol all testing sessions and the four supplemental testing sessions included exercising on a cycle ergometer at 50-60% of their max power output for 30 min. Following the bout, all participants performed the Thorstensson test on an isokinetic dynamometer for torque, power, and fatigue rate of the dominate leg quadriceps. **RESULTS:** The acute supplement x time interactions were not significant (p>0.05) for peak power (PL BL 469±81 W, PL D1 490±97 W *vs.* CM BL 465±85 W, CM D1 480±103 W), peak torque (PL BL 150±26 Nm, PL D1 157±32 Nm *vs.* CM BL 149±26 Nm, CM D1 156±33 Nm), fatigue rate (PL BL 57±9%, PL D1 57±10% *vs.* CM BL 57±10%, CM D1 56±9%), and heart rate (PL BL 156±17 bpm, PL D1 146±13 bpm *vs.* CM BL 155±11 bpm, CM D1 146±11 bpm). The chronic supplement x time interactions were not significant (p>0.05) for peak power (PL BL 469±81 W, PL D2 501±99 W *vs.* CM BL 464±85 W, CM D2 501±81 W), peak torque (PL BL 150±26 Nm, PL D2 161±31 Nm *vs.* CM BL 149±27 Nm, CM D2 161±26 Nm), fatigue rate (PL BL 57±9%, PL D2 58±9% *vs.* CM BL 57±10%, CM D2 58±9%), and heart rate (PL BL 156±17 bpm, PL D2 146±9 bpm *vs.* CM BL 155±11 bpm, CM D2 146±9 bpm). **CONCLUSION:** The results of this study suggest that acute and chronic supplementation of CM has no effect on recovery and fatigue rate of the quadriceps. Based on the data collected there was no significant differences between the recorded values for torque and power for each participant.

INTRODUCTION

Citrullline malate (CM) is a commonly sold over the counter ergogenic aid that has been gaining popularity due to its proposed ability to aid in reducing fatigue by increasing blood flow (1). Citrulline malate is composed generally in the ratio of 2:1 of citrulline, a nonessential amino acid, and malate, a Krebs cycle intermediate, respectively (2, 3). A majority of studies have suggested consuming citrulline and malate together with the above ratio increases the effectiveness because of the combined ability to enhance oxidative energy turnover, improve acid base balance, and lower energy cost for muscular force production (4, 5). The exact mechanism of how CM alleviates fatigue are not understood. One proposed theory is that CM is able to facilitate the clearance of ammonia and lactate through its role in the urea cycle. Additionally CM is thought to increase nitric oxide concentrations through the nitric oxide synthase- dependent pathway by converting L-citrulline into L-arginine via the intestinal-renal axis of the kidneys (3, 6) . Once the conversion occurs, L-arginine can then be converted into nitric oxide to aid in vasodilation and oxygen delivery. The production of NO has been shown to enhance recovery processes, exercise performance, mitochondrial respiration, muscle contractility, muscle repair, sarcoplasmic reticulum calcium handling, and glucose uptake (2, 7, 8). Supplementing with L-citrulline is thought to be more effective than supplementing with L-arginine because unlike L-arginine, L-citrulline is able to bypass hepatic metabolism and be directly transported to the kidneys for metabolism (7). This process has been shown to result in more than 80% of citrulline being recycled to endothelial cells for NO production (4, 8). Lastly, malate encourages oxidative energy turnover through its roles as a Krebs cycle intermediate, which may behave as a metabolic shuttle between the cytoplasm and mitochondria to help increase the rate of adenosine triphosphate production (4, 7). Citrulline Malate's effect on NO production, Krebs cycle functioning, and byproduct clearance suggests that it could have a significant effect on aerobic metabolism while alleviating a rapid onset of fatigue. Although the results on effectiveness of acute supplementation are mixed, it has been proposed that regular consumption may promote a more significant ergogenic benefit.

PURPOSE

The purpose of this study was to investigate the effects of acute and chronic citrulline malate supplementation on muscle fatigue among healthy male participants.

METHODS

IRB Approval: The study was approved by the Institutional Review Board for Human Subjects at Texas A&M University-Kingsville

Subjects: All subjects provided informed consent prior to testing. Eighteen (N=18) recreationally trained males (24±5 yr, 83±14 kg, 174±6 cm) were recruited from the student population at Texas A&M University-Kingsville and from fitness gyms within city of Kingsville.

METHODS, cont.

Pre-participation Screening/Testing: All subjects underwent health screening according to the American College of Sport Medicine's guidelines for exercise testing and prescription. Only subjects cleared to engage in moderate-to-vigorous intensity exercise based on these guidelines were allowed to participate. Tests for body composition (using equation for BMI), resting heart rate, resting blood pressure, maximal wattage on the stationary bike, and familiarization with the Biodex dynamometer were also conducted during the first laboratory visit. Testing for maximal wattage was done by having the participant perform a graded exercise test (GXT) on a cycle ergometer. The GXT began with a 5-min warm-up on the stationary cycle at a resistance of 50 watts. Following the warm-up, participants began pedaling at a cadence of 100 rpms while increasing the resistance by 25 watts every minute. This continued until participants were unable to maintain a cadence of 100 rpms. The highest wattage achieved for an entire minute was determined as their maximal power output.

Experimental Design: In a randomized, counterbalanced order, participants consumed a placebo (PL) and citrulline malate (CM) treatment for 2 separate dosing periods. There were a total of 6 experimental testing sessions, which included 2 baseline testing sessions and 4 supplemental testing sessions. For each dosing period, participants reported on 3 separate sessions with 7 days between each session. The first experimental testing session for each dosing period was considered the baseline day (BL), the second session the acute day (D1), and the third session the chronic day (D2). For chronic supplementation, all participants consumed each treatment for 7 consecutive days. For all experimental testing sessions, all participants cycled on standard cycle ergometer for 30 min. Following the 30 min cycling, all participants performed the Thorstensson isokinetic leg extension test (THOR).

Experimental Testing Procedures: All participants began by performing a standardized warm-up for 5 min on a stationary bike at 50 W. Following the warm-up, participants began cycling for 30 min above 70 rpms at 50-60% of each participant's max wattage from the GXT. Heart rate was measured every 5 min throughout the cycling period as a measure of intensity of activity, and averaged for data analysis. Upon completion of the cycle set, participants cooled down for 5 min by pedaling at a decreased wattage of 25W. Following the 5 min recovery period, participants performed the THOR test.

Isokinetic Leg Extension Test: Participants performed the THOR test on a Biodex dynamometer to determine changes in power, torque, and rate of fatigue within the quadriceps. Each participant performed 50 maximal leg extensions via the dominant leg at a constant rate of 180·sec^{-1}**.** Upon completion of the test, the results for the three highest and the three lowest values for power, torque, and fatigue rate from the 50 reps were recorded, and averaged, respectively, for data analysis.

Supplementation Procedures: The CM treatment consisted of 8 g of citrulline malate mixed into 20oz of sugar free flavored water. The Placebo (PL) treatment contained 20 oz of sugar free flavored water alone. The baseline testing sessions involved the participants consuming no beverage. For the 4 supplemental testing sessions, participants first consumed either the PL or CM drink, then began the exercise protocol one hour after finishing the drink to allow adequate digestion. Following the first supplemental testing day for both treatments, participants reported to the Human Performance Laboratory daily to consume their beverage (with the exception of weekends and other special circumstances in which they were given bags of the supplement to consume on their own).

Data Analysis: Two-way (supplement x time) ANOVAs with repeated measures were used to analyze for differences in the dependent variables between trials (PL & CM) across time (Baseline & D1 for acute supplementation; Baseline & D2 for chronic supplementation). If needed, appropriate *post-hoc* tests were used to make all pairwise comparisons for specific differences across the four experimental trials. The experimentwise error rate ($\alpha = 0.05$) was maintained throughout all *post hoc* tests for specific differences.

RESULTS

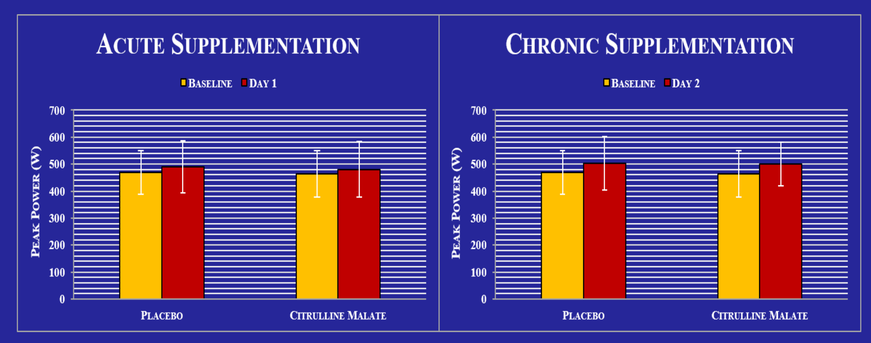

Figure 1: Supplement x Time Interaction for Peak Power. The change in peak power from BL to D1(p=0.759), and from BL to D2 (p=0.818) did not differ between supplements.

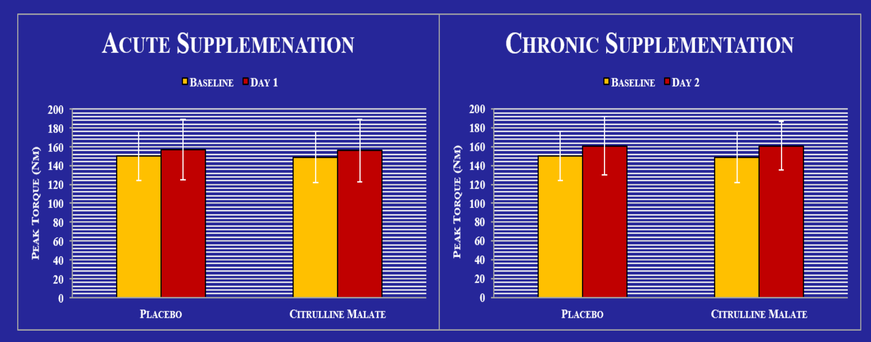

Figure 2: Supplement x Time Interaction for Peak Torque. The change in peak torque from BL to D1(p=0.940), and from BL to D2 (p=0.829) did not differ between supplements.

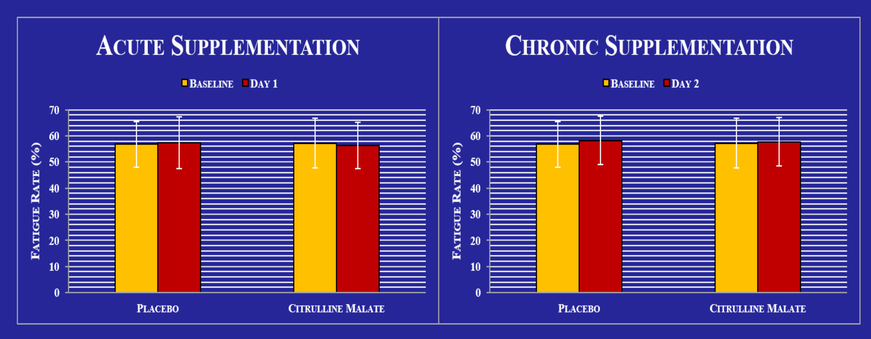

Figure 3: Supplement x Time Interaction for Fatigue Rate. The change in fatigue rate from BL to D1(p=0.631), and from BL to D2 (p=0.723) did not differ between supplements.

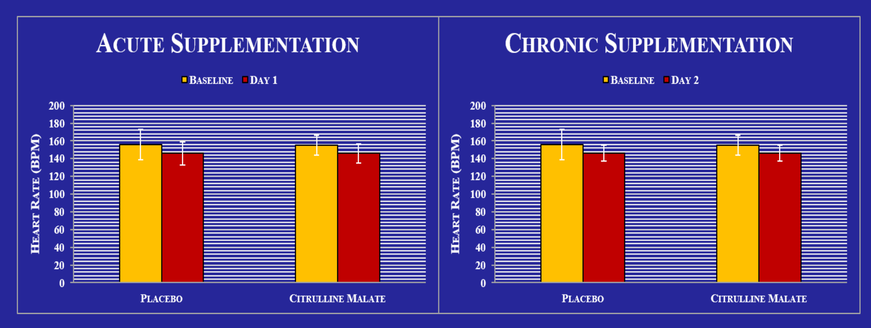

Figure 4: Supplement x Time Interaction for Heart Rate. The change in heart rate from BL to D1(p=0.754), and from BL to D2 (p=0.961) did not differ between supplements.

CONCLUSIONS

The results of this study suggest that acute and chronic supplementation of CM had no impact on recovery or fatigue rate of the quadriceps following 30 min of continuous cycling. There were no statistically significant differences within the quadriceps when measuring for torque, power, work or fatigue rate while performing maximal voluntary knee extensions on the Biodex dynamometer. To our knowledge, this investigation was the first to investigate the effects of both an acute and chronic supplementation of CM. Our protocol may not have induced a significant metabolic stimulus for citrulline malate to become effective in order to help with reducing muscle fatigue. Therefore, future research should incorporate protocols that promote a high metabolic demand while consuming CM chronically.

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