

Review

Effect of citrulline on post-exercise rating of perceived exertion, muscle soreness, and blood lactate levels: A systematic review and meta-analysis

Hye Chang Rhim ^{a,†}, Sung Jong Kim ^{a,†}, Jewel Park ^a, Ki-Mo Jang ^{a,b,*}

^a College of Medicine, Korea University, Seoul 02842, Republic of Korea

^b Department of Orthopaedic Surgery, Anam Hospital, Korea University College of Medicine, Seoul 02841, Republic of Korea

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Abstract

Background: Citrulline is one of the non-essential amino acids that is thought to improve exercise performance and reduce post-exercise muscle soreness. We conducted a systematic review and meta-analysis to determine the effect of citrulline supplements on the post-exercise rating of perceived exertion (RPE), muscle soreness, and blood lactate levels.

Methods: A random effects model was used to calculate the effect sizes due to the high variability in the study design and study populations of the articles included. A systematic search of PubMed, Web of Science, and ClinicalTrials.gov was performed. Eligibility for study inclusion was limited to studies that were randomized controlled trials involving healthy individuals and that investigated the acute effect of citrulline supplements on RPE, muscle soreness, and blood lactate levels. The supplementation time frame was limited to 2 h before exercise. The types and number of participants, types of exercise tests performed, supplementation protocols for L-citrulline or citrulline malate, and primary (RPE and muscle soreness) and secondary (blood lactate level) study outcomes were extracted from the identified studies.

Results: The analysis included 13 eligible articles including a total of 206 participants. The most frequent dosage used in the studies was 8 g of citrulline malate. Citrulline supplementation significantly reduced RPE ($n = 7, p = 0.03$) and muscle soreness 24-h and 48-h after post-exercise ($n = 7, p = 0.04$; $n = 6, p = 0.25$, respectively). However, citrulline supplementation did not significantly reduce muscle soreness 72-h post-exercise ($n = 4, p = 0.62$) or lower blood lactate levels ($n = 8, p = 0.17$).

Conclusion: Citrulline supplements significantly reduced post-exercise RPE and muscle soreness without affecting blood lactate levels.

Keywords: Amino acids; Dietary supplements; Ergogenic aid; Nitric oxide; Watermelon juice

1. Introduction

L-citrulline is one of the non-essential amino acids mainly found in watermelon (*Citrullus vulgaris*).¹ Supplements containing L-citrulline have been manufactured in different forms, including pure L-citrulline, watermelon juice, and citrulline malate (CM), and their effects have been studied under clinical and applied exercise settings.^{2–14} Recently, Trexler et al.¹⁵ published a meta-analysis evaluating the acute effect of citrulline supplementation on high-intensity strength and power performance. According to the meta-analysis, citrulline supplementation significantly enhanced the performance of high-intensity and power exercises.¹⁵ Other recent studies have investigated the effect of citrulline supplements on the

rating of perceived exertion (RPE) and muscle soreness, as well as exercise performance.^{2–14}

The potential use of citrulline supplements to relieve fatigue or muscle soreness is based on 2 hypothetical mechanisms. First, L-citrulline, one of the amino acids involved in the ureagenesis cycle, may facilitate the clearance of ammonia. Ammonia plays an important role in fatigue because its intracellular accumulation favors glycolysis while inhibiting the aerobic utilization of pyruvate.^{11,16} This modified energy metabolism results in the formation of lactate, which may contribute to fatigue.^{17,18} During high-intensity exercise, the rate of glycolysis is increased, and anaerobic glycolysis also leads to accumulation of blood lactate.¹⁸ The synthesis of lactate may be diminished via L-citrulline supplementation because it buffers ammonia through the urea cycle, thereby enhancing the aerobic utilization of pyruvate.¹⁹ Second, L-citrulline in the kidney can be converted to L-arginine,²⁰ which is a substrate for nitric oxide synthase (NOS).²¹ Unlike L-arginine, L-citrulline

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* Corresponding author.

E-mail address: kimo98@hanmail.net (K.M. Jang).

† These two authors equally contributed to this work.

bypasses the intestinal or hepatic elimination.²² Oral L-citrulline supplementation can increase plasma L-arginine concentrations and amplify NO-dependent signaling in a dose-dependent manner.²³ Indeed, previous studies have shown that citrulline supplementation increased blood NO levels²⁴ and plasma arginine availability²⁵ for NO synthesis. NO is not only a powerful vasodilator, but also a regulator of multiple physiological functions of skeletal muscles, such as glucose uptake and oxidation, mitochondrial biogenesis, contractile functions, and muscle repair, via satellite cell activation and the release of myotrophic factors.^{26–28} Because NO is involved in muscle contractile function and repair, enhanced NO production through citrulline supplementation may decrease muscle soreness.

Although additional studies are required to elucidate the mechanisms of action underlying the efficacy of citrulline supplementation, citrulline supplements have attracted researchers and sports coaches who are constantly looking for effective ways to decrease fatigue and muscle soreness after exercise. Therefore, the primary aim of this meta-analysis and systematic review was to evaluate the effect of citrulline supplements on the post-exercise RPE and muscle soreness. Since lactate accumulation has been associated with muscle fatigue and soreness, the secondary outcome was to assess the effect of citrulline supplements on blood lactate levels after exercise.

2. Methods

2.1. Search strategy and quality assessment

A systematic literature search was performed using PubMed and Web of Science databases to identify articles published up to April 2019 that evaluated the effects of L-citrulline or CM on the post-exercise RPE, muscle soreness, and blood lactate levels. The literature search process followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines.²⁹ Various combinations of keywords such as “citrulline”, “exercise”, “strength”, “power”, “performance”, “athlete”, “muscle”, “fatigue”, “recovery”, “exertion”, “soreness”, “lactate”, and “lactic acid” were used with “AND” or “OR” commands. A manual search was also performed using reference lists of identified relevant articles to ensure that all appropriate studies were included in the analysis. After deleting duplicate articles, 2 researchers (HCR and SJK) individually assessed each study based on predefined inclusion and exclusion criteria, as described in Section 2.2, to select the eligible articles. The quality of each article was assessed based on the Cochrane risk of bias tool for randomized controlled trials.³⁰ Unpublished clinical trials were also searched using the term “citrulline” on ClinicalTrials.gov to identify any unpublished trials.

2.2. Study selection criteria

The study inclusion criteria were: (1) randomized controlled trials of healthy individuals; (2) studies that investigated the effect of L-citrulline or CM; (3) placebo-controlled and blinded trials; (4) studies published in English and

accessible online through PubMed and Web of Science search engines or unpublished studies with results reported at ClinicalTrials.gov; (5) reports of changes in RPE, muscle soreness, or blood lactate level as an outcome, regardless of the nature of the exercise; and (6) L-citrulline or CM ingestion within 2 h of the exercise program. The supplementation ingestion time frame was limited to 2 h before exercise because it is known that plasma L-arginine peaks after an oral dose of L-citrulline within 1 h to 2 h after ingestion.³¹ Hence, studies involving supplementation >2 h before exercise likely missed the effect of acute supplementation of citrulline on muscle fatigue or soreness and thus were excluded from our analysis.

2.3. Data extraction and outcome measures

The characteristics of the studies, such as types and number of participants, types of exercise tests performed, supplementation protocols for L-citrulline or CM, and primary (RPE and muscle soreness) or secondary (blood lactate level) outcomes were extracted manually for each article. The means and SD of the RPE, muscle soreness, and blood lactate levels for the citrulline supplementation group and the placebo group were determined from the original data included in the article. If the data were presented only graphically, they were read via WebPlotDigitizer.^{32,33} WebPlotDigitizer is a web-based program that allows conversion of graphical data to numerical data through manual plotting.³³ Although WebPlotDigitizer relies on manual plotting, the intercoder reliability is high. WebPlot-Digitizer has been used in previous systematic reviews and meta-analyses, including those pertaining to nutritional supplements.^{33–35}

2.4. Statistical analysis

The standardized mean difference was used as a measure of effect size. The standardized mean difference was calculated by dividing the difference between the mean outcome of the citrulline supplementation group and that of the placebo group by the pooled SD, reflecting the size of the effect of the citrulline intervention relative to the variability observed in each study. Due to the high variability in study designs and study populations across articles included in the analysis, a random effects meta-analysis was carried out using the calculated effect sizes. Cochrane Q and I^2 statistics were calculated to assess the heterogeneity among the articles. Outliers were identified through Cook's distance, and sensitivity analysis was carried out to examine the impact of outliers on the meta-analysis. Publication bias was assessed using the Begg's rank correlation test and the Egger's regression asymmetry test. All analyses were carried out using R-3.5.3 software (R Foundation for Statistical Computing, Vienna, Austria).

3. Results

3.1. Eligible studies and characteristics of included studies

The initial search of published articles yielded a total of 172 articles. After deleting the duplicate articles, 85 articles remained. Of these 85 articles, 60 irrelevant articles were

excluded based on their titles. Based on the study selection criteria, 11 studies identified in the search qualified for the meta-analysis. A total of 2 additional articles were identified through the review of reference lists of articles. No unpublished clinical trials were identified on ClinicalTrials.gov. Thus, 13 studies were included in our meta-analysis (Fig. 1). A total of 7 studies examined RPE immediately post-exercise. A total of 4 studies measured subjective muscle soreness 24 h, 48 h, and 72 h post-exercise, and 3 studies analyzed subjective muscle soreness 24 h and 48 h post-exercise. A total of 8 studies measured blood lactate levels before and immediately after exercise.

A total of 13 studies comprising 206 participants met the inclusion criteria.^{2–14} The sample size of the studies ranged from 7 to 41, with an average of 16 participants. All participants were young and healthy and were recreationally active or resistance trained. In terms of age, the majority of the participants were in their 20 s. The dosage of CM used in the studies ranged from 6 g to 12 g; for L-citrulline, the dosage ranged from 3 g to 6 g. The most frequently used dosage was 8 g of CM. The exercise protocols included resistance exercises,^{2,3,5–8,10,11,13,14} cycling,^{4,12} and running.⁹ The characteristics of each study, including the supplementation protocols, types of exercise, and reported side effects are summarized in Table 1.

3.2. Study quality and risk of bias assessment

Most studies were judged to have a low risk of bias for each component in the Cochrane risk of bias tool.³⁰ All studies were randomized, and 2 studies specifically used random number generators.^{2,3} A total of 7 studies specifically stated that persons unrelated to the study performed concealment.^{2–5,7,8,11} Only 2 studies were not double blinded.^{6,12} In the study by Farney et al.,⁶ no drinks were given to the control group, and only the

participants were blinded. Blinding was incomplete in another study in which the authors compared natural watermelon juice, watermelon juice enriched with L-citrulline, and a placebo drink composed of various fruit flavors without any citrulline content.¹² The participants in the study would have been able to differentiate the watermelon juice from the placebo drink. However, this study was still included in this meta-analysis because the comparison was made between the results of watermelon juice and watermelon juice enriched with L-citrulline and the participants would not have been able to differentiate between those 2 drinks.¹² Also, because watermelon juice that was not enriched with L-citrulline contained only 1.17 g of L-citrulline, the authors justified its use by stating that this watermelon juice could serve as better placebo than another drink that could be differentiated. Other studies had a low risk for performance bias in that they masked placebo treatments by appearance and taste; in particular, 4 studies controlled for taste and smell by having participants wear a nose clip when consuming the supplement.^{7,8,13,14}

The studies generally provided instructions for participants to avoid factors before the exercise sessions that might affect the results of the study, such as vigorous exercise, dietary supplements, alcohol, and caffeine. Only 2 studies^{2,3} instructed the participants to continue, instead of avoiding, their usual supplements, and 1 study⁶ did not provide any instructions regarding supplements. The studies generally had a low risk for attrition bias because there was no missing outcome data, and only 2 studies reported minimal attrition due to factors unrelated to the study.^{5,13} Other time commitments was given as the reason for attrition in 1 study,³ and 1 study did not report a reason for attrition.² Although none of the studies had prespecified protocols, there were no other evidence of reporting bias.

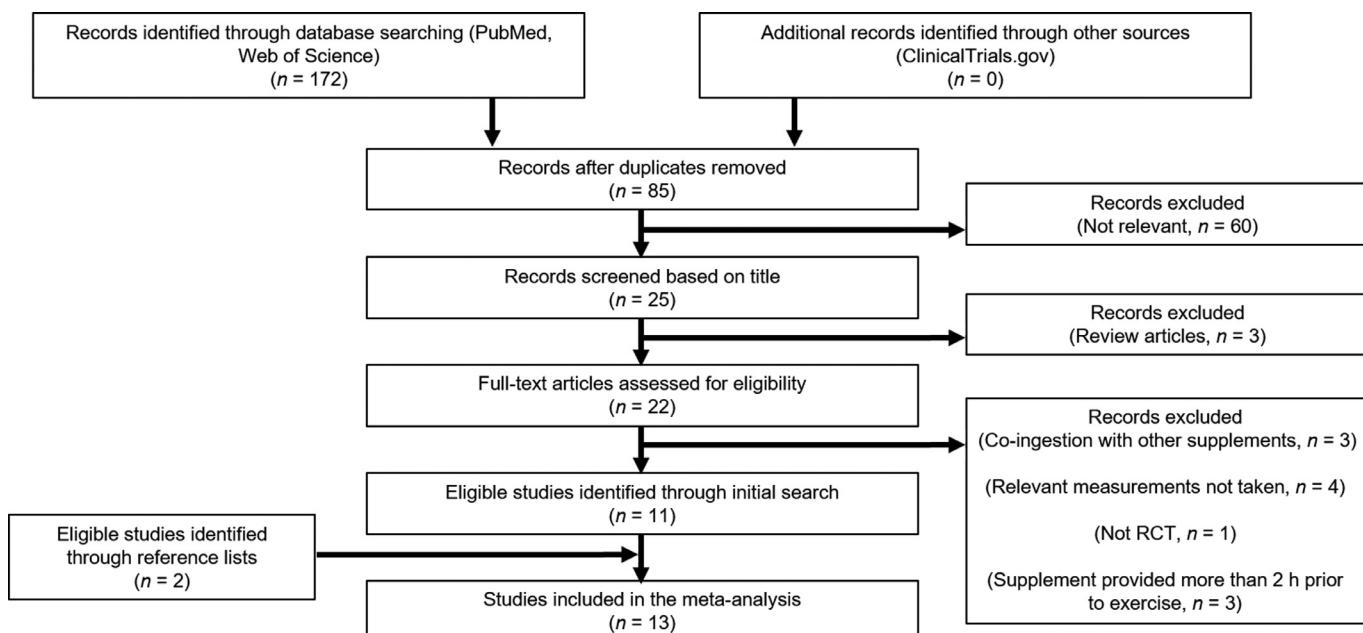


Fig. 1. Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) flow diagram of study selection process and results. RCT = randomized controlled trial.

Table 1

Characteristics of studies included in the analysis.

Study (first author, published year)	Design	Participants (<i>n</i> , age (mean \pm SD))	Supplement protocol	Exercise test	Primary and secondary outcomes	Side effects
1. Chappel et al. (2020) ²	RDB	Resistance-trained males and females (19, 25.7 \pm 7.7 years)	8 g CM 1 h before exercise	Ten sets of 10 repetitions of barbell curls at 80% of the participant's 1RM until failure with 60 s rest between sets	Muscle soreness, lactate level	No
2. Chappel et al. (2018) ³	RDB	Moderately trained males and females (15, 23.7 \pm 2.4 years)	8 g CM 1 h before exercise	Ten sets of 10 repetitions of single-leg extensor exercises at 70% of the subject's 1RM until failure with 60 s rest between sets	Muscle soreness, lactate level	No
3. Cunniffe et al. (2016) ⁴	RDB	Healthy trained males (10, 23.5 \pm 3.7 years)	12 g CM 1 h before exercise	Ten sets of 15 s maximal sprints on cycle ergometer with 30 s active recovery between sets	RPE, lactate level	No
4. da Silva et al. (2017) ⁵	RDB	Untrained males (9, 24.0 \pm 3.3 years)	6 g CM 1 h before exercise	Three sets of 8–12 repetitions of leg press and hack squats at 90% of 10 RM with 2 min rest between sets and exercises	RPE, muscle soreness, lactate level	No
5. Farney et al. (2019) ⁶	RSB	Recreationally trained males and females (12, 24.1 \pm 3.9 years)	8 g CM 1 h before exercise	Isokinetic leg extension test (15 maximal leg extensions) and 3 high-intensity exercise sessions (20 s each of squats, lunge jumps, squat jumps, and lateral jumps, with 30 s rest between exercises) with 1 min rest between rounds	Lactate level	N/A
6. Glenn et al. (2017) ⁷	RDB	Resistance-trained females (15, 23.0 \pm 3.0 years)	8 g CM 1 h before exercise	Six sets of bench press and leg press each at 80% of 1RM until failure with 60 s rest between sets and 120 s interval between exercises	RPE	No
7. Gonzalez et al. (2018) ⁸	RDB	Resistance-trained males (12, 21.4 \pm 1.6 years)	8 g CM 40 min before exercise	Five sets of 15 repetitions of bench press at 75% of 1RM until failure with 2 min rest between sets	RPE	N/A
8. Martínez-Sánchez et al. (2017) ⁹	RDB	Healthy amateur male runners (21, 35.3 \pm 11.4 years)	3.45 g C in WMJ 2 h before exercise	Squat jump and countermovement jump heights before and after 2 half-marathons	RPE, muscle soreness, lactate level	N/A
9. Martínez-Sánchez et al. (2017) ¹⁰	RDB	Resistance-trained males (19, 23.9 \pm 3.7 years)	3.3 g C in WMJ 1 h before exercise	Eight sets of 8 repetitions of half squats with 2 min rest between sets	RPE, muscle soreness	N/A
10. Pérez-Guisado et al. (2010) ¹¹	RDB	Healthy trained males (41, 29.80 \pm 7.64 years)	8 g CM 1 h before exercise	Four sets each of flat bench press (80% 1RM), incline bench press (80% 1RM for flat bench press), incline fly (60% 1RM for flat bench press), and flat bench press (80% 1RM) to failure with 1 min rest between sets and 2 min rest between exercises	Muscle soreness	Stomach discomfort after acute ingestion (<i>n</i> = 6)
11. Tarazona-Díaz et al. (2013) ¹²	RSB	Healthy males (7, 22.7 \pm 0.8 years)	6 g C in WMJ or 1.17 g C in WMJ 1 h before exercise	Eight repetitions of 30 s cycling with 4.5–5.0 kg resistance and 1 min rest between repetitions	RPE, muscle soreness	N/A
12. Wax et al. (2015) ¹³	RDB	Resistance-trained males (12, 22.1 \pm 1.4 years)	8 g CM 1 h before exercise	Five sets each of leg press, hack squats, and leg extension at 60% of 1RM with 3 min rest between sets	Lactate level	No
13. Wax et al. (2016) ¹⁴	RDB	Resistance-trained males (14, 23.3 \pm 1.5 years)	8 g CM 1 h before exercise	Three sets each of chin-ups, reverse chin-ups, and push-ups until failure with 3 min rest between sets	Lactate level	N/A

Abbreviations: C = citrulline; CM = citrulline malate; N/A = not applicable; RDB = randomized double-blinded; RM = repetition maximum; RPE = rating of perceived exertion; RSB = randomized single-blinded; WMJ = watermelon juice.

3.3. Meta-analysis of outcome measures

A meta-analysis of 7 studies investigating post-exercise RPE suggested that supplementation with L-citrulline or CM significantly reduced RPE, with an effect size of 0.81 ($p = 0.03$) (Fig. 2). A significant heterogeneity among the studies supported the choice of the random effects model ($I^2 = 81.9$, $Q = 28.9$, $p < 0.001$). Sensitivity analysis demonstrated that 1 study might have been highly influential, with a Cook's distance of 0.83, suggesting that this result may be an outlier.⁷ However, even after the exclusion of this study, citrulline supplementation resulted in a significant reduction in RPE ($p = 0.05$).

A meta-analysis of 7 studies investigating subjective feelings of muscle soreness 24-h post-exercise suggested that supplementation of L-citrulline or CM significantly reduced the feelings of muscle soreness, with an effect size of 0.99 ($p = 0.04$) (Fig. 3A). There was significant heterogeneity among the studies ($I^2 = 92.1$, $Q = 62.1$, $p < 0.001$), but no single study was found to be highly influential according to the sensitivity analysis.

No significant association ($p = 0.25$) was detected between citrulline ingestion and muscle soreness 48-h post-exercise (Fig. 3B). The result from 1 study was not included in the analysis due to identical values of muscle soreness in all subjects at 48 h after exercise in both citrulline supplementation and the placebo groups, and the standard deviation values of both groups were 0, making it impossible to calculate the effect size.¹² However, the majority of the studies suggested that citrulline supplements reduced muscle soreness at 48 h after exercise. Only one of the 6 studies suggested that there was a significant positive association between citrulline supplements and muscle soreness.³ A sensitivity analysis suggested that this study might have been highly influential, with a Cook's distance of 0.53, which may justify the exclusion of this study. After exclusion, a statistically significant association between citrulline supplements and muscle soreness 48-h post-exercise was observed ($p = 0.03$). There was significant heterogeneity among the studies ($I^2 = 93.4$, $Q = 73.7$, $p < 0.001$), which

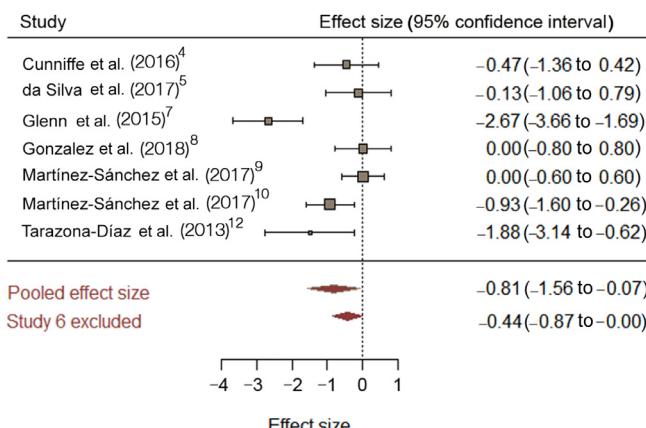


Fig. 2. The effect of citrulline supplementation on rating of perceived exertion.

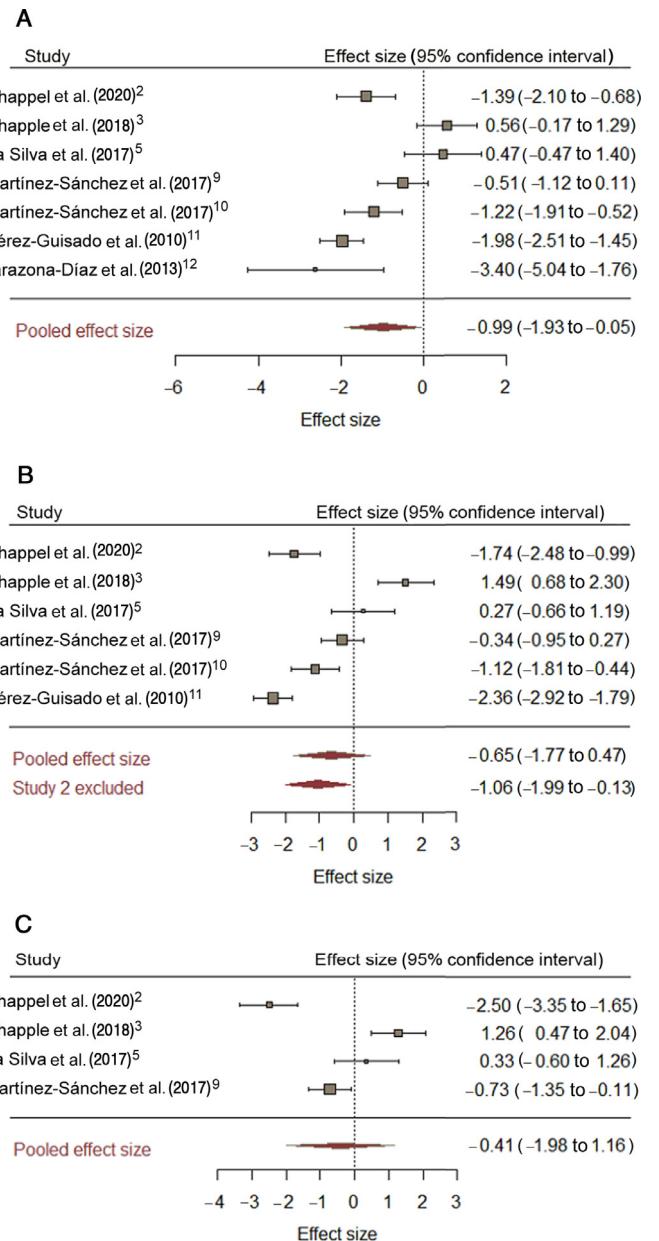


Fig. 3. The effect of citrulline supplementation on subjective rating of muscle soreness. (A) At 24 h after exercise; (B) At 48 h after exercise; (C) At 72 h after exercise.

remained significant even after exclusion of this study³ ($I^2 = 88.6$, $Q = 35.4$, $p < 0.001$). No significant association between citrulline ingestion and muscle soreness was observed at 72 h after exercise ($p = 0.62$) (Fig. 3C).

In a meta-analysis of 8 studies, citrulline supplements did not significantly reduce blood lactate levels after exercise ($p = 0.17$) (Fig. 4). No evidence of heterogeneity was observed ($I^2 = 0.0$, $Q = 6.6$, $p = 0.47$).

No significant asymmetries were observed in the funnel plots, and no evidence of publication bias was detected according to the Begg's rank correlation test and the Egger's regression test, in any of the analyses conducted.

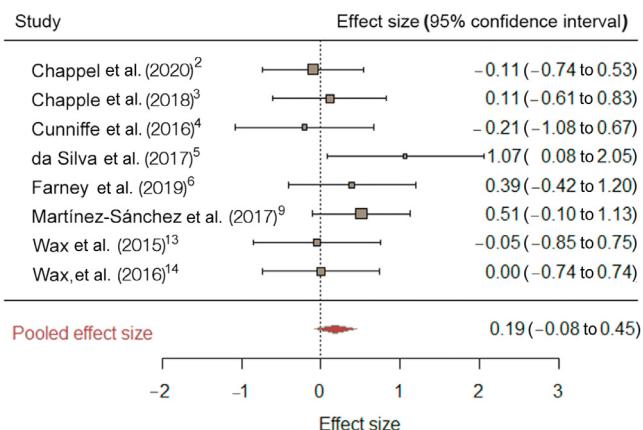


Fig. 4. The effect of citrulline supplementation on lactate levels after exercise.

4. Discussion

To the best of our knowledge, this systematic review and meta-analysis is the first to determine the effect of citrulline supplements on RPE, muscle soreness, and blood lactate levels. Our main finding was that citrulline supplements were effective in reducing both RPE and muscle soreness. The effect of citrulline on reducing muscle soreness was most significant at 24 h after exercise, and the effect appeared to diminish close to 72 h after exercise. This result may imply that the effect of citrulline occurred within 24 h of completing the exercise and declined subsequently. However, citrulline supplements did not significantly lower post-exercise blood lactate levels.

4.1. RPE and muscle soreness

All of the studies included in this analysis used either the Borg scale^{4,9,10,12} or OMNI (omnibus)-RPE scale,^{5,8} which are the 2 most widely used RPE tools.³⁶ The Borg scale ranges from 6 (*no exertion at all*) to 20 (*maximal exertion*),³⁶ and the OMNI-RPE ranges from 0 to 10 with mode-specific pictures.³⁷ These 2 scales have demonstrated reliability and validity in a healthy, clinical, and athletic adult population,³⁸ which is similar to the population included in this analysis. RPE can be used in both aerobic and anaerobic exercises to subjectively quantify individual perception of physical demands, exercise tolerance, and impending fatigue associated with activity.^{36,39} Additional recovery may be required for high RPE during or after exercise. In the absence of adequate recovery, a temporary reduction in muscle strength and increased feelings of soreness may occur,⁴⁰ which can last several days and affect the capacity to train at the desired intensity in the following sessions. The results of this meta-analysis suggest that either ingestion of CM or even L-citrulline alone may alleviate RPE and muscle soreness. Specifically, the effect size of citrulline supplements for RPE reduction was 0.81 and for reduction of muscle soreness at 24 h was 0.99, which may suggest a large difference between the citrulline supplementation group and the control group.⁴¹ However, due to the large heterogeneity in exercise protocols and the study population included in the analysis, it may not be

appropriate to discuss the magnitude of the effect size in this study.

4.2. Blood lactate levels

Only a few studies in animals and humans have suggested that citrulline supplementation may repress elevations of exercise-induced blood lactate levels. Using a swimming exercise protocol with mice, Takeda et al.⁴² demonstrated that L-citrulline supplementation inhibited the elevations in exercise-induced blood ammonia and lactate. Similar results were found in humans, showing that blood lactate levels were lower in a citrulline-supplemented group after running a half marathon.⁹ In that study, the authors also observed that L-citrulline ingestion increased the plasma concentration of lactate dehydrogenase (LDH), an enzyme known to reversibly convert pyruvate to lactate, with concomitant interconversion of nicotinamide adenine dinucleotide and its reduced form, nicotinamide adenine dinucleotide hydrogen. The study⁹ hypothesized that elevated plasma LDH might enhance the aerobic pathway even further by converting lactate to pyruvate.^{43–45} In mice, increased LDH gene expression exhibited enhanced oxygen consumption and mitochondrial enzyme activity,⁴⁶ but whether elevated plasma LDH can actually enhance the aerobic pathway still needs experimental evidence. In addition, none of the other studies included in our analysis found that citrulline supplementation attenuated blood lactate levels after exercise.^{2–6,13,14} This difference in lactate response may be attributed to differences in exercise or supplementation protocols. The studies that did not show a decrease in blood lactate levels involved anaerobic exercises, such as cycling sprint and time to exhaustion,⁴ and resistance exercises.^{2,3,5,6,13,14} By contrast, the study reporting attenuation used aerobic exercise (half marathon).⁹ Moreover, L-citrulline was supplemented in half marathon runners, whereas a CM supplement was given to subjects who performed anaerobic exercises.^{2–6,13,14} Therefore, malate may not provide additional benefit in lowering blood lactate levels, even though it is thought to bypass the ammonia-induced inhibition of the oxidative pathway and redirect lactic acid toward pyruvate production for aerobic utilization or gluconeogenesis.^{11,47} Interestingly, in 2 studies that were excluded from our analysis due to longer periods of supplementation (2 weeks⁴⁸ and 4 weeks⁴⁹), supplementation with CM attenuated lactate response after high-intensity exercise. Therefore, even though acute supplementation with CM may not be effective, CM supplementation for ≥2 weeks may lower blood lactate levels after high-intensity exercise.

The role of the blood lactate level has been challenged for its suitability as an indicator of muscle fatigue or soreness, since lactate accumulates in non-active muscles during exercise as well.⁵⁰ Rather than lowering blood lactate levels after exercise, citrulline supplements may act via other mechanisms to reduce the rating of fatigue and muscle soreness. One of the possible mechanisms may be based on energy production efficiency associated with the intake of citrulline supplements. Bendahan et al.⁵¹ showed that CM increased the rate of oxidative ATP production during exercise by 34% and the rate of

phosphocreatine recovery after exercise by 20%. In addition, Giannesini et al.⁵² found that short-term CM supplementation decreased the oxidative and phosphocreatine cost of skeletal muscle force generated in rat gastrocnemius muscles, indicating that CM supplementation may improve the efficiency of muscle contraction in rats. Bailey et al.⁵³ corroborated these results by demonstrating that citrulline supplementation enhanced oxygen uptake kinetics and improved tolerance during high-intensity exercise. Thus, efficient energy production during exercise and faster energy recovery after exercise may play a role in alleviating the sensations of fatigue and soreness.

4.3. Recommended dosage, timing, and safety

In studies reporting reduced RPE following citrulline supplementation, 8 g CM⁷ or watermelon juice containing 3.3 g¹⁰ and 6 g¹² of L-citrulline were used. Considering that the purported ratio of 2:1 L-citrulline to malate in CM may be as low as 1.1:1,² at least 3–4 g of L-citrulline alone or in CM may be necessary to alleviate fatigue. However, citrulline supplements were not effective in lowering RPE in other studies using 6 g CM,⁵ 8 g CM,⁸ 12 g CM,⁴ and 3.45 g L-citrulline in watermelon juice.⁹ This lack of response may be related to the timing of ingestion. In 5 studies,^{4,5,7,10,12} citrulline supplements were ingested 1 h before exercise to obtain the peak L-arginine levels after an acute oral dose of L-citrulline.³¹ In 2 studies reporting that citrulline supplements were not effective in reducing RPE, a supplement of 8 g CM was administered 40 min before exercise in 1 study,⁸ and 3.45 g L-citrulline in watermelon juice was provided 2 h before exercise in the other study.⁹ Thus, despite subtle differences, the timing of ingestion of citrulline supplements may be optimized by administering 1 h before exercise.

In terms of muscle soreness, there was no clear pattern or trend in which dosage and timing were effective. A total of 5 studies that demonstrated that citrulline supplements may reduce post-exercise muscle soreness used 8 g CM or 3–6 g L-citrulline.^{2,9–12} Also, supplementation of 3.45 g L-citrulline 2 h before a half marathon did not lower RPE but reduced post-exercise muscle soreness.⁹ Therefore, ingestion of citrulline supplements containing more than 3 g L-citrulline 1 h to 2 h before exercise may alleviate post-exercise muscle soreness.

Among the 13 studies, 8 reported the incidence of side effects. Although 6 studies stated that the participants did not experience any side effects, 1 study showed that 15% of the participants reported stomach discomfort after acute ingestion of 8 g CM.¹¹ One participant in another study reported gastrointestinal discomfort.⁷ However, it occurred after both CM and placebo trials. Thus, CM was unlikely to be the cause of self-reported discomfort.⁷ Supplementation with 12 g CM was well-tolerated by all 10 participants in 1 study.⁴ Therefore, CM ingested 1 h before exercise for the purpose of alleviating fatigue and muscle soreness is likely to be safe.

4.4. Limitations

Our study has some limitations. Although most studies used 8 g of CM containing 4–5 g of L-citrulline, the supplement dosages

differed slightly among the studies. Second, the exercise types varied among the studies (Table 1). A total of 9 studies implemented anaerobic exercises, with significant variation in the types of exercises, for example, bench press, leg press, and chin-ups. Also, only 1 study assessed the effect of L-citrulline in aerobic exercise. Such variations in the types of exercise might have contributed to the study heterogeneity, and the result of our analysis should be interpreted with caution when applied to aerobic exercise. In addition, the number of included studies was small because we limited studies to those that provided citrulline supplements 1 h or 2 h before exercise. Thus, studies in which supplements were given >2 h before exercise were excluded from this analysis. Because there were a limited number of studies evaluating the effect of long-term supplementation compared with acute supplementation, further research should investigate the effect of long-term supplementation on RPE and muscle soreness. Additional studies are required to elucidate the mechanisms by which citrulline supplements lower the perceived sensations of fatigue and muscle soreness.

5. Conclusion

The results of our systematic review and meta-analysis support the effectiveness of citrulline supplements in lowering RPE and muscle soreness. Athletes may benefit from ingesting either L-citrulline alone or CM 1 h before exercise to resist fatigue or relieve muscle soreness. Citrulline supplements are especially recommended for power and strength athletes for them to adequately recover and subsequently train at their desired intensity level. Further evidence is needed to confirm the efficacy of citrulline supplementation among endurance athletes.

Acknowledgments

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Authors' contributions

HCR conceived the study, designed it, and drafted the first version of manuscript; SJK performed the statistical analysis and drafted the first version of manuscript; JP drafted the first version of manuscript; and KMJ reviewed the study and drafted the final manuscript. All authors have read and approved the final version of the manuscript, and agree with the order of the presentation of the authors.

Competing interests

The authors declare that they have no competing interests.

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