Effects of L-Citrulline on Physical Performance

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Consumption of amino acids L-arginine (L-Arg) and L-citrulline (L-Cit) are purported to increase nitric oxide (NO) production and improve physical performance. Supplementing with L-Cit is more effective in increasing plasma L-Arg concentration than supplementing with L-Arg Clinical trials have shown relatively more favorable outcomes than not after supplementing with L-Cit and combined L-Arg and L-Cit. Administration of combined L-Arg and L-Cit may increase circulating L-Arg concentration and rapidly enhance NO bioavailability more effectively than either individual L-Arg or L-Cit, probably due to the synergistic effect of the two amino acids. Combining the two amino acids in supplements enhances performance by reducing energy expenditure, time exhaustion, and increases the power output.

exercise performance nitric oxide L-citrulline

1. Effects of L-Citrulline on Nitric Oxide Synthesis

While clinical studies examining supplementation effects across several different chronic conditions, including obesity, heart failure, and arterial stiffness, reported consistent significant increases in plasma nitrate and nitrite following acute or chronic L-Cit supplementation [1][2][3], the effect is inconsistent among healthy participants [4]. The increase in blood nitrate and nitrite concentration after L-Cit supplementation seems to be dose- and duration-dependent [5][6]. Consumption of 3.4 g/day L-Cit from watermelon juice for 16 days increased plasma L-Arg (116 ± 9 µM) compared to placebo (67 ± 13 µM) ($p < 0.001$) and plasma nitrite (201 ± 106 nM) compared to placebo (106 ± 21 nM) ($p < 0.05$) among recreationally active participants [5]. Consistent with these findings, consumption of 3 g/day L-Cit for seven days increased plasma nitrate/nitrite concentration on day 7 (35.2 ± 14.2 µM), compared to day 0 (22.4 ± 9.1 µM; $p < 0.05$) among track athletes [7]. However, most studies have demonstrated no changes in NO biomarkers in response to supplementation. For instance, Bailey et al. (2015), compared the effects of L-Cit 6 g/day against L-Arg 6 g/day and a placebo on NO biomarkers among recreationally active male participants [8]. Participants ingested 6 g L-Cit and 4.3 g maltodextrins, 6 g L-Arg and 4.3 g maltodextrins, or a placebo in 10.7 g maltodextrin for seven days. Following L-Cit supplementation, plasma L-Cit concentration significantly increased (665 ± 205 µM) compared to L-Arg (26 ± 6 µM) and the placebo (23 ± 5 µM; $p < 0.001$). Plasma L-Arg concentration significantly increased similarly in both L-Arg (151 ± 14 µM) and L-Cit (135 ± 22 µM) compared to the placebo (57 ± 14 µM; $p < 0.001$). Plasma nitrite concentration following L-Cit supplementation showed improvement yet did not significantly increase (100 ± 38 nM) compared to L-Arg (106 ± 41 nM; $p = 0.08$), and significantly increased after L-Arg supplementation (106 ± 41) compared to the placebo (83 ± 25 nM; $p < 0.05$).

Similarly, consumption of 2.4 g/day L-Cit for eight days significantly increased plasma L-Arg concentrations after cycling exercise protocol in the supplement group compared to the placebo group among trained male participants (192 ± 9 vs. 110 ± 4 nM/mL; $p < 0.001$) [9]. However, there was no difference in plasma nitrate and nitrite concentration in the supplement group compared to the placebo group ($p > 0.05$). Plasma NO was also not significantly different in the supplement group (58.4 ± 7.3 nmol/L^{-1}) compared to the placebo group (59.4 ± 13.6 nmol/L^{-1}) among swimmers, following eight days of supplementation with 8 g L-Arg or placebo ($p = 0.20$) [10]. A study by Hickner et al. (2006) reported a significant decrease in the plasma nitrate/nitrite concentration of the supplement group (21.18 ± 2.29) compared to the placebo group (24.58 ± 3.03 µM/L; $p < 0.05$) following ingestion of 3 or 9 g L-Cit and submaximal exercise protocol among recreationally active participants [11]. These findings contradict the reported efficacy of L-Cit to increase NO synthesis via increasing L-Arg bioavailability in healthy, active individuals.

2. Effects of L-Citrulline on Physical Performance and Perceptual Responses

An L-Cit dose of at least 3 to 4 g ingested 60 min before exercise is purported to be sufficient to alleviate fatigue [12]. Trexler et al. (2019) concluded that acute citrulline supplementation benefits high-intensity strength and power performance [13]. Based on the predetermined inclusion criteria, one article focused on the effect of L-Cit supplementation on resistance exercise performance. A study by Cutrufello et al. (2015) reported that a single dose of 6 g L-Cit consumed with a sucrose mixture 60 and 120 min before athletes completed 1 RM and graded exercise on a treadmill was not enough to improve exercise performance [14]. In that study, there were no significant changes in the number of repetitions during the chest press test (32.2 ± 11.4) compared to the placebo sucrose mixture (32.2 ± 10.5; $p = 0.51$). A study by Figueroa et al. (2017) reported that acute L-Cit does not enhance high-intensity exercise performance [15].

Similarly, ingestion of 2 g/day L-Cit for eight weeks combined with a strength training programme did not significantly improve bench press performance among resistance-trained male participants [1]. Unlike when co-ingested with malate, standalone L-Cit may not effectively increase strength performance. Potentially, the performance improvement may be related to malate's ability to increase the rate of ATP production and reduce lactate production during high-intensity anaerobic exercise [16].

Consumption of 6 g L-Cit before completion of a graded exercise test on a treadmill did not significantly change the time to exhaustion in the supplement group compared to the placebo (568 ± 96 vs. 559 ± 101 s; $p = 0.41$) and maximal oxygen consumption (4.0 ± 0.9 L/min vs. 4.0 ± 0.8 L/min; $p = 0.52$) [14]. A single dose of 6 g L-Cit may not be enough to improve aerobic or anaerobic performance among healthy, trained athletes [14][17]. However, 8 g of L-Cit consumed over eight days was not enough to improve 100 m or 200 m swimming time trial among swimmers [10]. A lack of endothelial dysfunction might explain the ineffectiveness of acute L-Cit supplementation among healthy young participants, who may benefit less from augmented NO synthesis [11][15]. L-Cit may be more effective when taken over several days than as a single bolus supplementation [18]. However, consuming 3.4 g/day L-Cit from watermelon juice concentrate over 16 days seems to have increased systolic blood pressure after supplementation compared to the control group (130 ± 11 vs. 124 ± 8 mmHg; $p < 0.05$). There were no differences in time to exhaustion for the supplement group compared to the placebo group during the severe intensity exercise test (550 ± 143 vs. 539 ± 108 s; $p > 0.05$), despite increased plasma nitrite concentrations following supplementation [5]. In contrast, consumption of 6 g/day L-Cit for four weeks significantly attenuated systolic blood pressure in the supplementation group compared to placebo (133 ± 5 vs. 139 ± 4; $p < 0.05$) among healthy young men [2]. Furthermore, other studies have demonstrated decreased systolic blood pressure following L-Cit supplementation [3][19][20].

Trained male cyclists consumed 6 g/day L-Cit or maltodextrin powder as a placebo for 7 days. On the final day, two hours after the last dose of the supplement, participants engaged in a cycling performance evaluation. In that study, time trial performance improved by 5.2% ($p = 0.08$), while average power output improved by 5.4% ($p < 0.05$) compared to the placebo [21]. Furthermore, the supplement group's average heart rate increased (159 ± 10 vs. 150 ± 13 bpm) and average RPE increased (16 ± 1 vs. 15 ± 1; $p < 0.05$) compared to the placebo. A similar dose of 6 g/day L-Cit lowered the mean arterial pressure in the supplementation group compared to the placebo (85 ± 2 vs. 87 ± 3 mmHg; $p < 0.05$) and reduced the volume of oxygen consumption mean response time (54 ± 5 vs. 60 ± 8 s; $p < 0.05$). At the same time, total work completed increased during the test for the supplementation group compared to the placebo (125 ± 19 vs. 123 ± 18 kJ; $p < 0.05$), and tolerance to severe intensity exercise significantly improved for the supplementation group compared to placebo (661 ± 107 vs. 589 ± 101 s; $p < 0.05$) among recreationally active participants [8]. Interestingly, plasma nitrate and nitrite only tended to be significant; suggesting that increased tolerance to severe intensity exercise may result from a different mechanism. As an amino acid intermediate, L-Cit reportedly plays an essential role in the urea cycle in reducing ammonia toxicity in the muscles. Takeda et al. (2011) reported that L-Cit supplementation attenuated exercise-induced blood ammonia levels during high intensity exercise in rabbits and in mice, and suggested that human participants may gain similar effects [22].

A dose of 3 g/day L-Cit for seven days reportedly decreased the RPE ($p = 0.015$), while mean power output ($p = 0.046$) and oxygen consumption ($p = 0.007$) increased after an intermittent short time duration high-intensity protocol for the L-Cit group compared to the placebo group among collegiate track athletes [7]. The effectiveness of L-Cit was also reported in response to a low dose of 2.4 g/day consumed for eight days by athletes; cycle ergometer time trial performance completion time was reduced by 1.5%, while mean power output was increased by 2% in the treatment group compared to the placebo group ($p < 0.05$) [9]. In that study, subjective feelings of fatigue and concentration also improved for the supplement group compared to the placebo. A study by Martínez-Sánchez et al. (2017) reported that L-Cit supplementation is vital to attenuate post-race muscle soreness and enhance aerobic pathways, resulting in lower plasma lactate concentrations during a half-marathon race [23]. The decreased plasma lactate was supported by reduced muscle glycogen utilisation during exercise, which suggested less reliance on anaerobic glycolysis for energy production [22]. L-Cit supplementation enhances oxidative production of ATP by inhibiting additional glycolysis by suppressing ammonia levels, thereby preventing activation of phosphofructokinase, which is a crucial marker for anaerobic glycolysis [23].

3. Effects of Combined L-Arginine and L-Citrulline Supplementation on Physiological, Perceptual Responses and Physical Performance

Few studies have assessed the efficacy of a combined L-Arg and L-Cit supplement on physiological responses and exercise performance [24]. Studies have combined L-Arg and L-Cit with BCAA at a ratio of 1:1 for L-Arg and L-Cit [25][26][27].

3.1. Effects of Combined L-Arginine and L-Citrulline on Nitric Oxide Synthesis

Male collegiate soccer players ingested 1.2 g L-Arg and 1.2 g L-Cit or placebo in granulated powder in a stick packet for seven days and then performed a cycling test [28]. In the supplement group, the plasma concentration of nitrite significantly increased post-exercise (45.2 ± 2.4 µM) compared to the placebo (37.8 ± 2.4 µM; $p < 0.05$). Supplementing with combined L-Arg and L-Cit reportedly demonstrates a synergistic effect [24][29]. L-Cit acts as a strong allosteric inhibitor, as it has an inhibiting effect on arginase, which metabolises L-Arg to urea and L-ornithine [30]. According to Khalaf et al. (2019), augmenting L-Arg as a substrate for NOS-3 maintains NO production, while L-Cit activates NOS-2 and indirectly activates NOS-1 in skeletal muscle leading to heightened NO synthesis [31]. However, a combination of 1 g L-Arg and 1 g L-Cit in a capsule for eight days was insufficient to increase plasma nitrate and nitrite concentration among healthy male participants, while plasma L-Arg concentrations reportedly increased [30]. This may be due to the ratio being 1:1 of L-Cit to L-Arg, instead of the recommended effective 2:1 ratio of L-Cit to L-Arg [24].

3.2. Effect of Combined L-Arginine and L-Citrulline on Physical Performance and Perceptual Responses

Ingestion of combined 1.2 g L-Arg and 1.2 g L-Cit or placebo in a granulated powder for seven days before completing a cycle ergometer test significantly increased power output (242 ± 24 vs. 231 ± 21 W; $p < 0.05$) and reduced subjective perception of ease of pedaling (5.8 ± 1.8 vs. 6.9 ± 19; $p < 0.05$) compared to the placebo among soccer players [28]. L-citrulline supplementation may also improve force production during anaerobic performance [32]. Similar effects on anaerobic performance are evident. Specifically, taekwondo athletes performed high-intensity intermittent exercise on a cycle ergometer to mimic taekwondo match physiological demands for 60 min following ingestion of amino acid pills or placebo [25]. Their premotor reaction time was significantly faster (0.139 ± 0.016) in the supplement group compared to the placebo (0.162 ± 0.010 s; $p < 0.001$). In a similar supplementation protocol, the supplement group of the high school swimmers was faster in the first lap (30.47 ± 0.47 s) compared to the placebo group (31.34 ± 0.57 s; $p < 0.001$) during a high-intensity interval swimming protocol.

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