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Article *in* Journal of Sports Sciences · November 2024

DOI: 10.1080/02640414.2024.2423578

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Journal of Sports Sciences

ISSN: (Print) (Online) Journal homepage: www.tandfonline.com/journals/rjsp20

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To cite this article: Emilia Zawieja, Steven Machek, Nelo Eidy Zanchi, Jason Cholewa & Małgorzata Woźniewicz (08 Nov 2024): Effects of chronic betaine supplementation on exercise performance: Systematic review and meta-analysis, Journal of Sports Sciences, DOI: 10.1080/02640414.2024.2423578

To link to this article: https://doi.org/10.1080/02640414.2024.2423578

JOURNAL OF SPORTS SCIENCES
https://doi.org/10.1080/02640414.2024.2423578

PHYSIOLOGY AND NUTRITION

Effects of chronic betaine supplementation on exercise performance: Systematic review and meta-analysis

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ABSTRACT
Betaine supplementation, a dietary practice that possesses potential effects on exercise performance, has undergone extensive study. This study aimed to systematically review and meta-analyse betaine supplementation's effects on exercise performance. We searched PubMed, Web of Science, Scopus, and Google Scholar, focusing on studies comparing chronic betaine to a placebo in healthy humans aged 15–60 years, measuring exercise outcomes. Studies with acute betaine supplementation, no control group, or animals were excluded. Quality assessment was done using the Cochrane Risk of Bias Tool, and a random-effects model was employed for the meta-analysis. The review included 17 studies with 317 participants (21% female). The results revealed a significant effect size of 0.47 (95% CI 0.04 to 0.89) for maximal strength (1RM, 3RM, maximal isokinetic or isometric force), particularly in the lower body (SMD: 0.49, 95% CI 0.01 to 0.98). No significant effects were found for upper body strength, cycling sprint power, bench press throws power, or muscular endurance. However, vertical jumping performance improved significantly (SMD: 0.36, 95% CI 0.03 to 0.69) after excluding a low-quality study. In conclusion, betaine supplementation for at least 7 days significantly enhances muscular strength, especially lower body strength, and shows potential in improving vertical jumping performance.

ARTICLE HISTORY
Received 9 December 2023
Accepted 23 October 2024

KEYWORDS
Betaine; muscular strength; muscular power; endurance; exercise; supplementation

1. Introduction

In the pursuit of optimizing athletic performance, athletes, coaches, and researchers are continuously exploring innovative strategies to enhance exercise capabilities and achieve competitive advantage. Among these strategies, nutritional supplementation has emerged as a focal point of investigation, with an array of compounds touted for their potential to augment physiological adaptations and improve exercise outcomes. One compound that has garnered considerable attention is betaine, a naturally occurring methyl donor found in various foods, including beets, spinach, and whole grains (Craig, 2004). Over the past two decades, betaine supplementation has gained prominence within the realm of sports nutrition due to its purported capacity to influence exercise performance (Cholewa et al., 2013; Hoffman et al., 2009; Zawieja et al., 2023)

Betaine, chemically known as trimethyl glycine, is implicated in a myriad of biological processes, including osmoregulation, cellular methylation, and the synthesis of key metabolite (Ueland, 2011). These multifaceted roles have led researchers to speculate on its potential impact on exercise-related mechanisms. Specifically, investigations into the effects of betaine supplementation on muscle strength, power, and endurance have sparked interest in the sports and exercise community.

Betaine's name derives from sugar beets, from where it was first synthesized in the 19th century (Cholewa et al., 2014). The first evidence of betaine as an ergogenic aid came in 1952 from poliomyelitis-diseased patients (Fallis & Lam, 1952). Betaine – guanidinoacetate supplementation in these patients enhanced general strength and endurance. However, the exact mechanisms of action are unknown. Although first used to increase lean mass and decreased fat mass in livestock such as pigs and chickens (Cholewa et al., 2014) humans have subsequently demonstrated similar body composition improvements (Cholewa et al., 2013).

A systematic review published in 2017 concluded that betaine supplementation did not enhance muscular strength or power (Ismaeel, 2017). At the time, only seven studies were available, and the authors could not perform a meta-analysis; however, nine additional studies have since been published (Arazi et al., 2022; Cholewa et al., 2018; Machek, Harris, et al., 2022; Moro et al., 2020; Nobari, Cholewa, et al., 2021; Waldman et al., 2023; Yang et al., 2020, 2022; Zawieja et al., 2023). Given the growing body of research in this domain, an updated systematic review and meta-analysis that critically evaluates the collective findings is crucial for a comprehensive understanding of the potential benefits and limitations of betaine supplementation in the context of exercise performance.

The proposed mechanisms by which betaine affects exercise performance include its impact on gene expression regulation by altering the methylation potential of cells, protection against metabolic or heat stress due to its osmoactive properties, improved recovery due to its antioxidant and anti-

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Supplemental data for this article can be accessed online https://doi.org/10.1080/02640414.2024.2423578

inflammatory action, and the upregulation of protein synthesis pathways through increased IGF-1 and testosterone concentrations (Apicella, 2011; Willingham, 2021; Zawieja et al., 2023). Although it is possible that betaine could deliver acute performance improvements by regulating cell hydration, our systematic review focused on the chronic effects resulting from the cumulative impact of betaine on muscular adaptations, including increased training volume, improved recovery, hormonal changes, and upregulation of gene expression – effects that can only be observed after repeated exposures.

The aim of this systematic review is to synthesize and analyse the existing literature on the effects of chronic betaine supplementation on exercise performance. The secondary aim was to analyse the effect of betaine on body composition and biochemical or hormonal markers.

2. Materials and methods

2.1. Eligibility criteria

The present systematic review followed the PRISMA statement (Page et al., 2021) and included clinical trials that determined the effects of betaine supplementation on exercise performance. The PICOS approach was used (Table 1).

We included both parallel group and cross-over studies with at least a 2-week washout period since it was previously shown that 2 weeks are sufficient to return serum betaine concentrations to baseline (Machek, Harris, et al., 2022; Machek, Zawieja, et al., 2022; Zawieja et al., 2023). No language, publication date, or publication status restrictions were imposed. We considered athletic performance the primary outcome after chronic (minimum one week) betaine intervention. The cut-off of one week was established as it represents the minimum time required to observe the initial chronic adaptations in gene expression, enzyme activity modulation, and sustained shifts in metabolic pathways. This cut-off is commonly used in supplementation studies. Acute effects of betaine (hours to days after ingestion) are primarily attributed to its osmoregulatory function and reflect direct impacts on performance, hydration status, and immediate biochemical changes. In this systematic review, however, we aimed to assess betaine's effects on chronic adaptations. The minimum betaine dose of 2 g/day was selected after a literature review indicated that no studies have assessed the effects of doses lower than 2 g/day on athletic performance. Other parameters than athletic performance were considered secondary outcomes in this systematic review. Exclusion criteria were as follows: acute betaine supplementation (<1 week), lack of control group, no exercise performance outcome, and animal model studies.

Table 1. PICOS criteria for inclusion of studies.

Parameter	Criteria
Participants	Trained or untrained, males or females, healthy, age 15-60 years
Intervention	Betaine supplementation for min. 7 days with a minimum dose of 2 g/d
Control	Identical placebo
Outcome	Exercise performance
Study types	Clinical trials with parallel and crossover design

2.2. Information sources

MEDLINE/PubMed, Web of Science, and Scopus databases were consulted in this review. Moreover, we used grey literature through Google Scholar. The last search was run on 7 August 2023. Additionally, we manually searched websites in the area, using the same keywords. No publication date filter was applied in any search.

2.3. Search strategy

MeSH, Entry Terms, and keywords related to betaine and exercise performance were used. Full MEDLINE search strategy was adapted for the other databases. Grey literature search was performed using the same search terms in Google Scholar. There were no language restrictions, although English search terms were used. Study-type restrictions were not applied during the search. The full search strategy is described in Supplementary Material 1.

2.4. Study selection

Eligibility assessment was performed independently, in a duplicate standardized manner by two reviewers (EZ and SM). Disagreements were resolved by discussion between the two review authors; if no agreement could be reached, it was resolved by the third (NEZ) author.

2.5. Data collection process

Data from the included studies were extracted by one review author (EZ) and checked by a second author (SM). Disagreements were resolved by discussion between the two review authors; if no agreement could be reached, it was resolved by a third author (NEZ). Synthesis of the results included the following: authors and year; country; study type; size and characteristics of the sample; intervention (dose, duration); placebo group; exercise performed; primary outcome; secondary outcome. Quantitative analysis was performed for the following outcomes: vertical jump (VJ), maximal muscular strength (upper and lower body), muscular endurance, power in the bench press (BP) throw, and cycling sprint (peak and mean power). Where data were incomplete, authors were contacted to obtain the relevant information. Graphed data was extracted using WebPlotDigitizer 4.0 (https://automeris.io/WebPlotDigitizer, Pacifica, CA, USA, accessed: August 2023)

2.6. Quality assessment (risk for bias)

To ascertain the validity of eligible clinical trials, a pair of reviewers (EZ and MW), working independently and with adequate reliability, determined the quality of included studies using the Cochrane Risk of Bias 2 tool for parallel and cross-over designs (Higgins et al., 2003). After grading the included studies, EZ and MW discussed the discrepancies to reach a consensus. If an agreement could not be reached, a third researcher, NEZ, would make the final decision. Publication bias was detected visually with funnel plots.

2.7. Certainty of evidence

Certainty of evidence was assessed using the Grades of Recommendation, Assessment, Development and Evaluation (GRADE) system with GRADEpro GDT: GRADEpro Guideline Development Tool [Software], McMaster University and Evidence Prime, 2023 (Available from gradepro.org).

2.8. Meta-analysis calculations

We used the standardized mean difference (SMD) as effect size. For muscular strength and endurance, we performed a subgroup analysis of upper and lower body strength. Descriptive analyses were performed using a spreadsheet (Microsoft Excel 2016© USA), whereas meta-analytic statistics were made with Review Manager (RevMan) version 5.3 (Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2014). SMD, the number of participants, and the standard error of the SMD for each study were used to quantify changes in the performance variables when comparing betaine ingestion vs. placebo. Considering that the effect of betaine on performance may differ according to dose and other moderators relating to participants, we decided to use a random effects model. The Cohen criteria were used to interpret the magnitude of SMD: <0.2, trivial; 0.2–0.5, small; 0.5–0.8, moderate; and >0.8, large (Cohen, 2013). For heterogeneity analysis, we calculated the I (Hoffman et al., 2009) statistic, which indicated the percentage of observed total variation across studies due to real heterogeneity. An I (Hoffman et al., 2009) value between 25% and 50% represents a small amount of inconsistency, an I (Hoffman et al., 2009) value between 50% and 75% represents a medium amount of heterogeneity, and an I (Hoffman et al., 2009) value >75% represents a large amount of heterogeneity (Higgins et al., 2003). To ensure outcome independence, multiple outcomes from a single study were pooled. Meta-regression was conducted for outcomes with at least eight independent studies. The moderators included the duration of supplementation, age, training status (untrained, recreationally trained, trained), sex (male, female, mixed), and exercise type. Meta-regression was performed using R Statistical Software version 4.4.1 (R Foundation for Statistical Computing, Vienna, Austria) with the metafor and meta packages.

3. Results

Utilizing our search strategy, we identified 794 studies within MEDLINE, 3173 within Web of Science, 654 within Scopus, and 1190 within Google Scholar. After eliminating duplicates, 2069 studies were left. Among these, 2046 studies were excluded after reviewing the abstracts as they did not fulfill the eligibility criteria. Out of the 23 remaining studies, 6 did not meet the specified inclusion criteria (two studies used acute betaine supplementation) (Armstrong et al., 2008; Dantas et al., 2021) three studies used multi-ingredient supplementation (Kraemer et al., 2007; Schwarz et al., 2019; William Kedia et al., 2014) and no exercise test was performed in the other one (Willingham, 2021). Consequently, 17 studies aligned with the inclusion criteria and were incorporated into the systematic review (Apicella, 2011; Arazi et al., 2022; Cholewa et al., 2013, 2018; Del Favero et al., 2012; Hoffman et al., 2009, 2011; Lee et al., 2010; Machek, Harris, et al., 2022; Moro et al., 2020; Nobari, Cholewa, et al., 2021; Pryor et al., 2012; Trepanowski et al., 2011; Waldman et al., 2023; Yang et al., 2020, 2022; Zawieja et al., 2023). The process of final study selection is depicted in the PRISMA flow diagram (Figure 1). Sixteen studies were published in peer-reviewed journals, while one study was a master's thesis (Apicella, 2011).

3.1. Participants and study general characteristics

The included studies spanned from 2009 to 2023. Eight studies were published from 2009 to 2013. The remaining nine studies were published from 2018 to 2023. No studies were published during the period between 2014 and 2017. Among the included studies, one was conducted in the following countries: Brazil, Italy, and Poland (Table 2). Two studies were conducted in Iran and two in Taiwan. Furthermore, 10 studies originated from the United States. Of these, two studies do not clearly state whether they were randomized (Yang et al., 2020, 2022) while 15 were randomized clinical trials. All the chosen studies followed a double-blinded and placebo-controlled approach. Eight studies utilized a cross-over design, while the remaining nine employed parallel groups.

The sample sizes varied, ranging from 10 to 43 participants, resulting in a total of 317 participants with a median of 17. Among these participants, 79% were males ($n = 250$), whereas 21% were females ($n = 67$). Among the included studies, only two involved solely female participants (Cholewa et al., 2018; Waldman et al., 2023) and two included both males and females (Moro et al., 2020; Pryor et al., 2012). The remaining 13 studies involved only male participants. Regarding the training status of participants, nine studies included recreationally trained individuals, two studies included untrained individuals, two studies focused on CrossFit enthusiasts, and one study each involved participants engaged in handball, martial arts, and soccer training. Notably, one study did not provide information about the training status of its participants (Yang et al., 2020). The mean age of the overall sample was 23.5 years. It is worth mentioning that one study only provided the age range without reporting the mean age (Del Favero et al., 2012). The mean body mass across the studies was 76.9 kg, and the mean height was 174.4 cm. Notably, three studies omitted reporting height information (Cholewa et al., 2013; Del Favero et al., 2012; Lee et al., 2010). The general characteristics of each study can be found in Table 1.

3.2. Intervention characteristics

Intervention characteristics can be found in Table 2. Fifteen studies solely focused on betaine supplementation. One study evaluated not only betaine supplementation but also the impact of blood flow restriction (Machek, Harris, et al., 2022). Another study assessed the effects of betaine, creatine, and combined betaine+creatine supplementation (Del Favero et al., 2012). For the purpose of data extraction in this systematic review, we exclusively considered the results from the betaine-alone group and the placebo group of the del Favero study (Del Favero et al., 2012). Similarly, in the study by Machek,

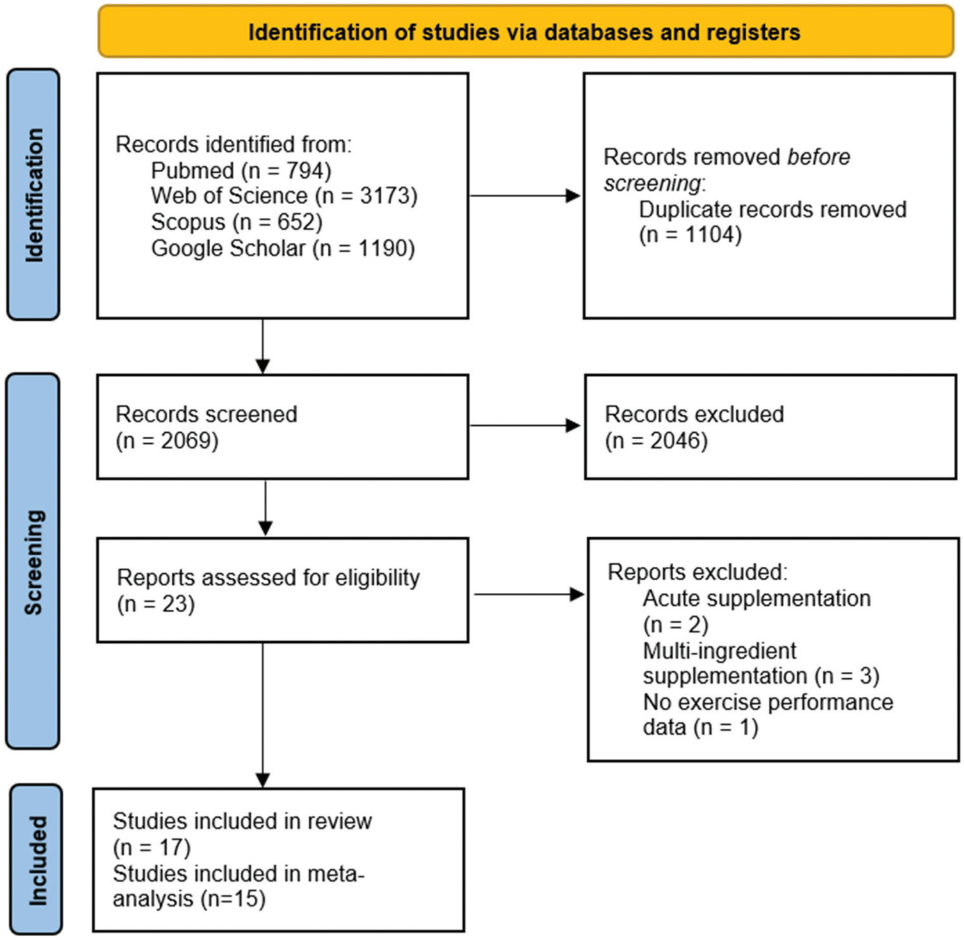

Figure 1. Flow diagram of study identification by CONSORT.

Harris, et al. (2022) we isolated the effects of betaine supplementation, disregarding the effects of blood flow restriction. Furthermore, from the study by Lee et al. (2010) we solely extracted data from day 1 of testing (baseline and after supplementation). From the Nobari, Cholewa, et al. (2021) study, we exclusively considered data from the baseline and week 14 time points. Lastly, for the Hoffman et al. (2009) study, we exclusively utilized data from the baseline and day 14-time points Studies' characteristics are presented in Table 2.

In studies with a parallel design, the allocation of participants was balanced, with 89 participants in both the betaine group and the placebo group. However, Hoffman et al. (2009) did not provide information about group allocation, and for the purposes of this meta-analysis, we assumed equal allocation. Betaine doses ranged from 2.0 to 6.0 g/d, with the most used dose being 2.5 g (used in 13 studies). Betaine was administered in various forms: dissolved in a sports drink (in 7 studies), dissolved in water (in 3 studies), and encapsulated (in 7 studies). Placebo was administered in the same forms as betaine: either as a sports drink alone (in 7 studies), mixed with water (using maltodextrin, dextrose, or microcrystalline cellulose) (in 3 studies), or as inactive placebo capsules containing substances like flour, sugar, cellulose, microcrystalline cellulose, and carboxymethyl cellulose (in 7 studies). The duration of supplementation varied from 7 to 98 days, with a mean duration of 26.2 ± 23.4 days (median: 14 days).

3.3. Quality assessment

The quality assessment results are presented in Supplementary Material 2. Among cross-over studies, six were ranked as having "some concerns", one as "low risk of bias", and another one as "high risk of bias". Among parallel-group studies, seven were ranked as "some concerns", one as "low risk of bias", and another one as "high risk of bias". The two studies were most susceptible to bias because they do not clearly state whether they were randomized (Yang et al., 2020, 2022).

3.4. Certainty of evidence

The summary table of certainty of evidence is available in Supplementary Material 3. Three outcomes (vertical jump, cycling peak power and cycling average power) were rated as "high", one outcome (muscular strength) was rated as "moderate" and two outcomes (muscular endurance and bench press throw) as "low" certainty.

Table 2. Study characteristics.

Author, year	Country	Study design	N total	N males	N females	Training status	Mean age	Mean BM	Mean height
Apicella et al. (2011)	United States	RCT, DB, PL-CON, CR, 14-day	12	12	0	recreationally active	19.7	84.3	172.6
Arazi, 2022	Iran	RCT, DB, PL-CON, CR, 30-day washout	10	10	0	trained, handball,	16	78.8	182
Cholewa, 2013	United States	RCT, DB, PL-CON, PG	23	23	0	recreationally resistance trained,	25	72	NI
Cholewa, 2018	United States	RCT, DB, PL-CON, PG	33	0	33	untrained	20.9	68.6	165.9
del Favero, 2012	Brazil	RCT, DB, PL-CON, PG	17	17	0	untrained	NI	69	NI
Hoffman, 2009	United States	RCT, DB, PL-CON, PG	24	24	0	recreationally active	20.9	80.6	179.1
Hoffman, 2011	United States	RCT, DB, PL-CON, CR, 4-week washout	11	11	0	recreationally, resistance-trained	21.7	79.8	178.5
Lee, 2010	United States	RCT, DB, PL-CON, CR, 14-day washout	12	12	0	recreationally, resistance-trained	21	79.1	NI
Machek, 2022	United States	RCT, DB, PL-CON, PG	18	18	0	recreationally, resistance-trained	25	87.6	177.9
Moro, 2020	Italy	RCT, DB, PL-CON, PG	29	15	14	trained Crossfit	34.9	70.7	171.2
Nobari, 2021	Iran	RCT, DB, PL-CON, PG	29	29	0	trained soccer	15.5	62.5	173.2
Pryor, 2012	United States	RCT, DB, PL-CON, CR, 21-day washout	16	9	7	recreationally active	19	75	172
Trepanowski, 2011	United States	RCT, DB, PL-CON, CR 21-day washout	13	13	0	recreationally, resistance-trained	23	82	178
Waldman, 2023	United States	RCT, DB, PL-CON, PG	23	0	23	recreationally active,	21.8	66.6	166
Yang, 2020	Taiwan	nsRCT, DB, PL-CON, CR, 3-week washout	10	10	0	NI	24.6	76.5	177
Yang, 2022	Taiwan	nsRCT, DB, PL-CON, PG	16	16	0	trained martial arts,	20	69	175.7
Zawieja, 2023	Poland	RCT, DB, PL-CON, CR, 21-day washout	43	43	0	trained Crossfit	34.1	82.3	178.6

Abbreviation list: BM, body mass; CR, cross-over; DB, double-blinded; NI, not informed; nsRCT, not stated if randomized clinical trial; PG, parallel groups; PL-CON, placebo-controlled; RCT, randomized clinical trial.

3.5. Performance tests

Included studies used a variety of exercise performance tests. VJ was measured in eight studies (Cholewa et al., 2013, 2018; Hoffman et al., 2009; Lee et al., 2010; Nobari, Cholewa, et al., 2021; Trepanowski et al., 2011; Yang et al., 2022); one repetition maximum (1RM) in BP in 5 studies (Cholewa et al., 2013, 2018; Del Favero et al., 2012; Nobari, Cholewa, et al., 2021; Yang et al., 2022); 1RM in squat (SQ) in 4 studies (Cholewa et al., 2013, 2018; Del Favero et al., 2012; Yang et al., 2022); BP endurance in four studies (Arazi et al., 2022; Hoffman et al., 2009; Lee et al., 2010; Trepanowski et al., 2011); and cycling sprints in four studies (Hoffman et al., 2009; Pryor et al., 2012; Waldman et al., 2023; Zawieja et al., 2023); isometric BP (Apicella, 2011; Lee et al., 2010; Trepanowski et al., 2011); and BP throw (Hoffman et al., 2009; Lee et al., 2010; Trepanowski et al., 2011) in 3 studies each; 1RM in leg press (LP) (Machek, Harris, et al., 2022; Nobari, Cholewa, et al., 2021); isometric SQ (Apicella, 2011; Lee et al., 2010); LP endurance (Arazi et al., 2022; Machek, Harris, et al., 2022); SQ endurance (Hoffman et al., 2009; Lee et al., 2010) in 2 studies each. The remaining performance tests were used in only one study each (power output in 6 rep x 60% 1RM in BP and SQ) (Del Favero et al., 2012) isokinetic peak concentric (CON) and eccentric (ECC) force in chest press (Hoffman et al., 2011) isokinetic muscle endurance in chest press 5 sets x 6 reps x 80% of peak CON and ECC force (Hoffman et al., 2011) 3 reps SQ jump (Lee et al., 2010) 3RM in SQ, 2 km rowing, Bergeron beep test (Moro et al., 2020) change of direction, 10 m sprint, 30 m sprint, repeated sprints, 30–15 intermittent fitness test (for VO_2max) (Nobari, Cholewa, et al., 2021) isometric force in LP (Trepanowski et al., 2011) graded exercise test (Waldman et al., 2023) exhaustive endurance exercise (Yang et al., 2020) overhead Medicine-Ball Throw Test, 1RM in overhead press, sumo deadlift (Yang et al., 2022) and Fight Gone Bad (Zawieja et al., 2023).

3.6. Quantitative analysis of primary outcomes

In the quantitative analysis of maximal muscular strength, nine studies were included, resulting in 17 outcomes (8 for upper body strength and 9 for lower body strength). The following outcomes were considered: 1RM in BP (Cholewa et al., 2013, 2018; Del Favero et al., 2012; Nobari, Cholewa, et al., 2021; Yang et al., 2022) 1RM in SQ (Cholewa et al., 2013, 2018; Del Favero et al., 2012; Yang et al., 2022) 1RM in LP (Machek, Harris, et al., 2022; Nobari, Cholewa, et al., 2021) 1RM in overhead press and sumo deadlift (Yang et al., 2022) 3RM in SQ (Moro et al., 2020) maximal ICC and ECC force in chest press (Hoffman et al., 2011) and maximum isometric BP and SQ (Trepanowski et al., 2011). Multiple outcomes from the same study were pooled in order to ensure the independence of effect sizes (Hoffman et al., 2011; Machek, Harris, et al., 2022; Yang et al., 2022).

The results of the meta-analysis revealed a significant difference ($p = 0.03$) between betaine and placebo trials concerning measures of maximal strength (Figure 2). The pooled SMD for the effects of betaine ingestion on muscle strength was 0.47 (95% confidence intervals (CI): 0.04, 0.89, medium heterogeneity I = 78%). Subgroup analysis indicated that betaine significantly improve lower body strength (SMD = 0.49; 95% CI: 0.01, 0.98; $p = 0.05$; medium heterogeneity I = 69%) but does not have a significant effect on upper body strength (SMD = 0.50; 95% CI: −0.25, 1.25; $p = 0.19$; high heterogeneity I = 84%).

A sensitivity analysis demonstrated that the result of the meta-analysis for upper body strength did not alter after excluding single studies. Only after excluding Cholewa et al. (2013), the upper body strength was close to reaching significance (SMD: 0.70, 95%CI: −0.10, 1.51, $p = 0.09$). Sensitivity analysis also revealed that excluding Cholewa et al. (2018) (SMD: 0.50, 95% CI: −0.05, 1.05, $p = 0.07$), Nobari, Cholewa, et al. (2021) (SMD: 0.49, 95% CI: −0.06, 1.04, $p = 0.08$), Trepanowski et al. (2011) (SMD: 0.49, 95% CI: −0.06, 1.04, $p = 0.08$), or Yang et al. (2022) (SMD: 0.50, 95% CI: −0.06, 1.06, $p = 0.08$) from the analysis of lower body strength resulted in the loss of a significant effect of betaine on lower body strength. However, this exclusion did not impact total-body strength, which remained significantly greater with betaine supplementation.

The meta-regression analysis was performed separately for upper-body strength (moderators: duration of supplementation, age, sex, and training status) and lower-body strength (moderators: duration of supplementation, age, sex, training status, and exercise type (squat vs. leg press)) (Supplementary Material 4). The results of the meta-regressions did not show any significant relationships between the moderators (duration of supplementation, age, sex, training status, and exercise type) and strength outcomes.

The qualitative analysis of muscular power was categorized into VJ (7 studies) (Cholewa et al., 2013, 2018; Hoffman et al., 2009; Lee et al., 2010; Nobari, Cholewa, et al., 2021; Trepanowski et al., 2011; Yang et al., 2022), sprint cycling (4 studies) (Hoffman et al., 2011; Pryor et al., 2012; Waldman et al., 2023; Zawieja et al., 2023), and BP throw power (3 studies) (Hoffman et al., 2009; Lee et al., 2010; Trepanowski et al., 2011). The results of the meta-analysis showed no significant differences between betaine and placebo for any of the muscular power outcomes ($p > 0.05$; heterogeneity: low I = 4% for VJ, low I = 0% for sprint cycling peak power, low I = 0% for sprint cycling average power and high I = 91% for the bench press throw) (Figures 3 and 4). A sensitivity analysis indicated that only when Yang et al. (2022) was excluded, VJ performance showed significant improvement with betaine compared to placebo (SMD = 0.36; 95% CI: 0.03, 0.69; $p = 0.03$). However, sensitivity analysis did not reveal any changes in sprint cycling or bench press throw power.

Six studies with 9 outcomes were included in the meta-analysis of muscular endurance (Arazi et al., 2022; Hoffman et al., 2009, 2011; Lee et al., 2010; Machek, Harris, et al., 2022; Trepanowski et al., 2011). In case of multiple outcomes from a single study, the data were pooled (Hoffman et al., 2011; Machek, Harris, et al., 2022). The types of endurance tests are presented in Table 3. The meta-analysis results did not reveal any significant effect of betaine on muscular endurance compared to placebo (SMD = 0.43; 95% CI: −0.36, 1.23; $p = 0.29$, high heterogeneity I = 87%) (Figure 5). Subgroup analysis also did not show any differences between betaine and placebo for upper and lower body endurance (upper body: SMD = 0.15; 95% CI: −1.10, 1.40; $p = 0.82$, high heterogeneity I = 90%; lower body: SMD = 0.79; 95% CI: −0.25, 1.82; $p = 0.14$, high heterogeneity I = 84%). Sensitivity analysis showed that the results of muscular endurance did not change (total, upper, and lower body) after excluding any of the studies.

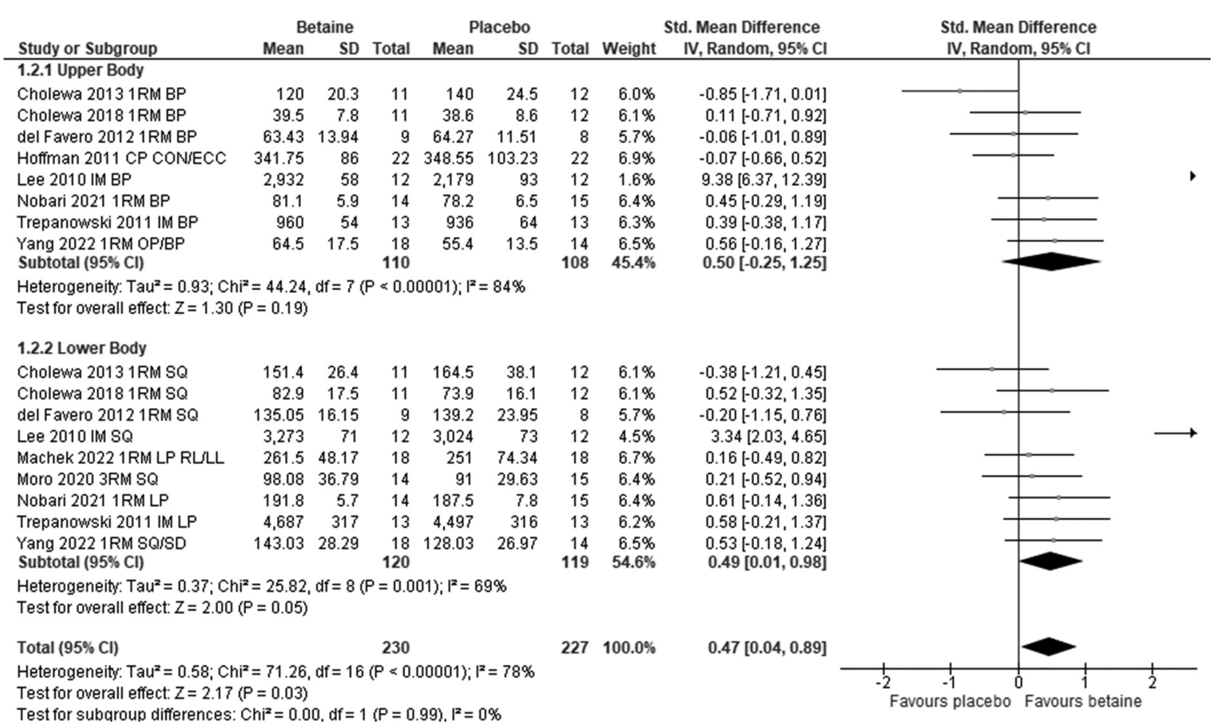

Figure 2. Meta-analysis results of muscular strength. Abbreviation list: 1RM, one repetition maximum; 3RM, three repetitions maximum; BP, bench press; CON, concentric force; CP, chest press; ECC, eccentric; IK, isokinetic; IM, isometric; LL, left leg; LP, leg press; OP, overhead press; RL, right leg; SD, sumo deadlift; SQ, squat.

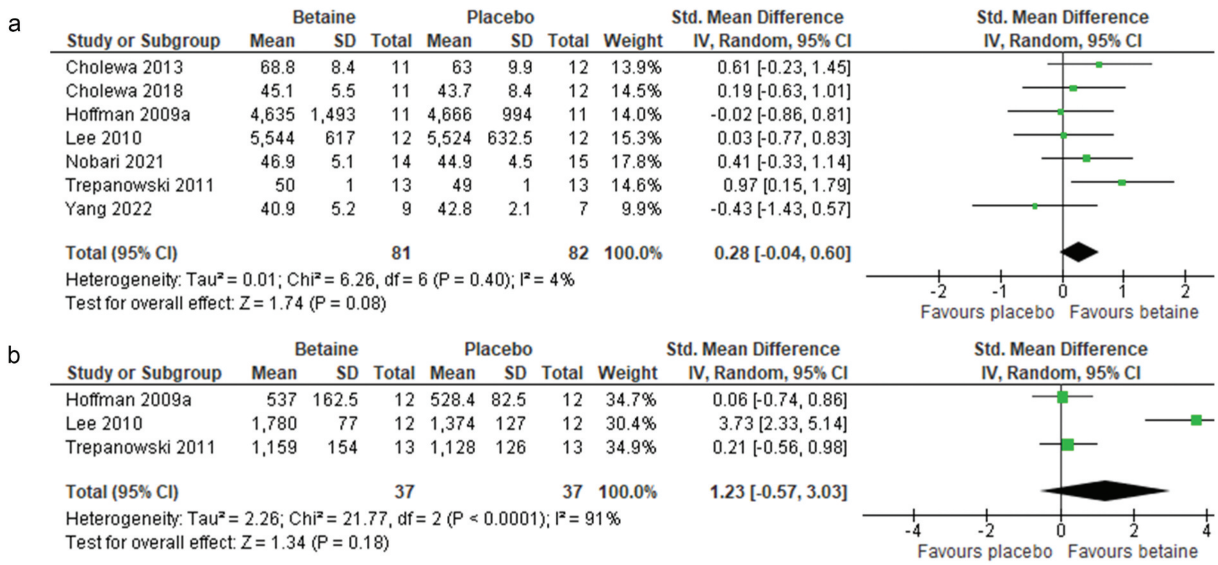

Figure 3. Meta-analysis results of vertical jump (panel A) and bench press throw (panel B).

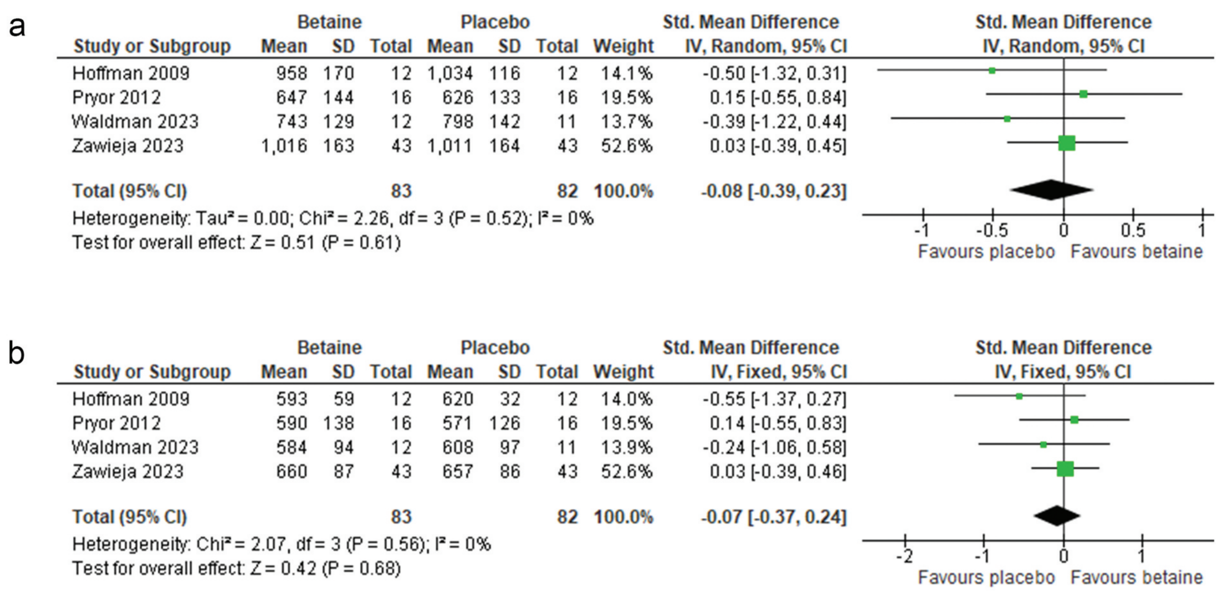

Figure 4. Meta-analysis results of cycling peak power (panel A) and average power (panel B).

3.7. Qualitative assessment of secondary outcomes

Body composition was assessed in five of the included studies (Cholewa et al., 2013, 2018; Moro et al., 2020; Waldman et al., 2023; Zawieja et al., 2023). Betaine supplementation resulted in increased fat-free mass compared to baseline in two studies (Cholewa et al., 2013; Waldman et al., 2023) and compared to placebo in one study (Cholewa et al., 2018). Body fat and fat mass decreased following betaine supplementation compared to baseline in only one study (Cholewa et al., 2013) and compared to placebo in another study (Cholewa et al., 2018). Total body water, intracellular water, and extracellular water were not affected by betaine.

Regarding hormone concentrations, insulin-like growth factor-1 (IGF-1) (Apicella, 2011; Machek, Harris, et al., 2022; Zawieja et al., 2023) and cortisol (Apicella, 2011; Arazi et al., 2022; Zawieja et al., 2023) were measured in three studies each, while growth hormone (Apicella, 2011; Machek, Harris, et al., 2022) and testosterone (Arazi et al., 2022; Zawieja et al., 2023) were measured in two studies each. IGF-1 (Machek et al., 2022; Apicella 2011) and total testosterone (Apicella, 2011; Zawieja et al., 2023) concentrations increased after betaine supplementation compared to baseline in two studies. Growth hormone concentrations increased in only one study (Apicella, 2011) and cortisol concentrations decreased with betaine in two studies (Apicella, 2011; Arazi et al., 2022).

Lactate was measured in five studies (Arazi et al., 2022; Lee et al., 2010; Machek, Harris, et al., 2022; Trepanowski et al., 2011; Waldman et al., 2023) but a significant decrease after betaine supplementation was observed only in one of them (Arazi et al.,

Table 3. Intervention characteristics.

Author	Betaine dose	PL	Duration	Type of performance test	Primary outcomes (performnce)	Secondary outcomes (body composition, hormonal, biochemical, immunochemical)
Apicella, 2011	2.5 g/d powder mixed with sport drink	sport drink	14 days	VJ, isometric BP, isometric SQ, box lifting	VJ, isometric BP: difference with betaine vs. difference with placebo ↔ Isometric SQ, box lifting: difference with betaine vs. difference with placebo ↑	GH, IGF-1: before betaine vs. after betaine ↑, betaine vs. placebo ↔ Cortisol: before betaine vs. after betaine ↓, betaine vs. placebo ↔
Arazi, 2022	2.5 g/d powder mixed with water	maltodextrine powder mixed with water	14 days	LP and BP: 5 sets to failure 80% 1RM	LP and BP reps: betaine vs. placebo ↑	Total testosterone, testosterone to cortisol: before betaine vs. after betaine: ↑, betaine vs. placebo: ↑ Cortisol, lactate: before betaine vs. after betaine: ↓, betaine vs. placebo: ↓
Cholewa, 2013	2.5 g/d capsules	flour capsules	6 weeks	VJ, 1RM BP and SQ	VJ, 1 RM BP, 1 RM SQ: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔	Thigh CSA: before betaine vs. after betaine ↔, betaine vs. placebo ↔ Arm CSA, LBM: before betaine vs. after betaine ↑, betaine vs. placebo ↔ BF, FM: before betaine vs. after betaine ↓, betaine vs. placebo ↔
Cholewa, 2018	2.5 g/d capsules	sugar capsules	8 weeks	VJ, 1RM BP and SQ	VJ, 1 RM BP, 1 RM SQ: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔	BF, FM: betaine vs. placebo: ↓ FFM: betaine vs. placebo: ↑ TBW, ICW, ECW, Muscle Thickness: betaine vs. placebo: ↔
del Favero, 2012	2 g/d powder mixed with water	dextrose powder mixed with water	10 days	Power in 6 rep x 60% 1RM BP and SQ and 1 RM BP and SQ	Average power BP and SQ, 1RM BP and SQ: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔	Muscle PCr: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔
Hoffman, 2011	2.5 g/d powder mixed with sport drink	sport drink	15 days	Isokinetic peak CON and ECC force in CP, isokinetic muscle endurance in CP 5 sets x 6 reps x 80% of peak CON and ECC force	Isokinetic CON and ECC peak force, CON and ECC endurance: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔	
Hoffman, 2011	2.5 g/d powder mixed with sport drink	sport drink	14 days	VJ, BP throw z 30% 1RM, 75% 1RM reps to failure in SQ and BP, 2 × 30s x 1.2 Nm·kg-1 resistance WAnT	BP and SQ reps to failure, WAnT, BP throw, VJ: betaine vs. placebo: ↔	
Lee, 2010	2.5 g/d powder mixed with sport drink	sport drink	14 days	4 sets × 3 reps VJ, isometric SQ, 3 reps SJ 30% 1RM, 3 sets x 85% 1RM to failure in SQ, isometric BP, 3 reps BP throw x 30% 1RM, 3 sets x 85% 1RM to failure in BP	VJ (2 out of 4 jumps), isometric S force: before betaine vs. after betaine: ↑, betaine vs. placebo: ↔ SJ, SQ and BP to failure: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔ Isometric BP force, BP throw power: before betaine vs. after betaine: ↑, betaine vs. placebo: ↑	Glucose, haematocrit, haemoglobin, plasma osmolality, lactate: betaine vs. placebo: ↔
Machek, 2022	6 g/day capsules	cellulose capsules	14 days	1RM left/right leg LP and LP repetitions to failure	1RM LP left and right leg, reps to failure LP: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔	Lactate, GH, HIF-1A: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔ IGF-1: before betaine vs. after betaine: ↑, betaine vs. placebo: ↔
Moro, 2020	2.5 g/d powder mixed with water	microcrystalline cellulose powder mixed with water	6 weeks	3 RM SQ, 2 km rowing, Bergeron beep test	3 RM SQ: before betaine vs. after betaine: ↑, betaine vs. placebo: ↔ 2 km rowing, Bergeron beep test: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔	FM, FFM, TBW: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔

(*Continued*)

Table 3. (Continued).

Author	Betaine dose	PL	Duration	Type of performance test	Primary outcomes (performnce)	Secondary outcomes (body composition, hormonal, biochemical, immunochemical)
Nobari, 2021	2 g/d capsules (flour)	flour capsules	14 weeks	VJ, change of direction, 10 m sprint, 30 m sprint, repeated sprints, 30-15 intermittent fitness test (for VO_2max), 1RM BP and LP	VJ, 1RM LP and BP, change of direction, repeated sprint peak power: before betaine vs. after betaine ↑, difference with betaine vs. placebo ↑ Change of direction, 10 m sprint, repeated sprint min power: before betaine vs. after betaine ↔, difference with betaine vs. placebo ↔ 30 m sprint: before betaine vs. after betaine ↓ VO_2max: before betaine vs. after betaine ↑	
Pryor, 2012	2.5 g/d mixed with sport drink	sport drink	7 days	4 × 12 s x 5.5% BM resistance cycloergometer sprints	Average and maximum peak power, average and maximum mean power: betaine vs. placebo: ↑, before betaine vs. after betaine: ↑	
Trepanowski, 2011	2.5 g/d powder mixed with sport drink	sport drink	14 days	VJ, BP throw x 30% 1RM, isometric force in LP, isometric force in BP, 10 sets x 10 reps x 50% 1RM in BP	VJ, BP throw, isometric LP and BP, BP total reps and volume: before betaine vs. after betaine: ↔, betaine vs placebo: ↔	Lactate, nitrate, malondialdehyde, upper torso circumference: before and after betaine: ↔, betaine vs. placebo: ↔
Waldman,2023	2.4 g/d capsules	micro-cellulose capsules	2 weeks	3 × 10s x 7.5% BM resistance cycloergometer sprints, GXT cycling	Mean power sprint no. 3: before betaine vs. after betaine: ↑, betaine vs. placebo: ↔ Mean power sprint 1,2, Peak power sprint 1,2,3; HR, RPE (three first stages): before betaine vs. after betaine: ↔, betaine vs. placebo: ↔ RPE (fourth stage): before betaine vs. after betaine: ↓, betaine vs. placebo: ↔	BF, FM, lactate: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔ FFM: before betaine vs. after betaine: ↑, betaine vs. placebo: ↔ CHO oxidation: before betaine vs. after betaine: ↑, betaine vs. placebo: ↓ fat oxidation: before betaine vs. after betaine: ↓, betaine vs. placebo: ↔
Yang, 2020	2.5 g/d powder mixed with sport drink	sport drink	14 days	Exhaustive endurance exercise	TTE, VO_2peak: betaine vs. placebo: ↔	Betaine concentration: before betaine vs. after betaine: ↑, betaine vs. placebo: ↑ Choline concentration: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔
Yang, 2022	2.5 g/d capsules	carboxymethyl cellulose capsules	6 weeks	Overhead Medicine-Ball Throw Test, VJ, 1RM BP, overhead press, half SQ, sumo dealift	Overhead Medicine-Ball Throw, 1RM BP, overhead press, half SQ and sume deadlift: before betaine vs. after betaine: ↑, betaine vs. placebo: ↔ VJ: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔	TG, HDL-C, LDL-C: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔ TC: before betaine vs. after betaine: ↓, betaine vs. placebo: ↔
Zawieja, 2023	2.5 g/d or 5 g/d capsules	cellulose capsules	21 days	Figh Gone Bad (3 rounds: wall ball, sumo deadlift high pull, box jump, push press, rowing), 1 × 30s x 7.5% BM WAnT	FGB total reps: before betaine vs. after betaine: ↑, betaine vs. placebo: ↑ WAnT peak power, mean power and minimum power: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔	FM, FFM, TBW, IGF-1, cortisol: before betaine vs. after betaine: ↔, betaine vs. placebo: ↔ Testosterone: before betaine vs. after betaine: ↑, betaine vs. placebo: ↔

Abbreviation list: 1RM, one repetition maximum; BM, body mass; BP, BF, body fat; bench press; CHO, carbohydrate; CON, concentric; CP, chest press; CSA, cross sectional area; ECW, extracellular water; EXC, eccentric; FFM, fat-free mass; FM, fat mass; GH, growth hormone; GXT, graded exercise test; HDL-C, high density lipoprotein cholesterol; HIF-1A, hypoxia-induced factor 1 alpha; ICW, intracellular water; IGF-1, insulin-like growth factor 1; LDL-C, low density lipoprotein cholesterol; LP, leg press; PCr, phosphocreatine; SJ, squat jump; SQ, squat; TBW, total body water; TC, total cholesterol; TG, triglycerides; TTE, time to exhaustion; VJ, vertical jump; VO_2max, maximum oxygen uptake, WAnT, Wingate test.

2022). Muscle phosphocreatine was measured in a single study, with no differences observed with betaine supplementation (Del Favero et al., 2012).

Glucose, haematocrit, haemoglobin, plasma osmolality, muscle hypoxia-inducible factor-1 alpha expression, nitrate, malondialdehyde, choline, triglycerides, high-density lipoprotein cholesterol, and low-density lipoprotein cholesterol were each measured in a single study, and no differences were observed with betaine (Lee et al., 2010; Machek, Harris, et al., 2022; Trepanowski et al., 2011; Waldman et al., 2023; Yang et al., 2020). Total cholesterol was measured in a single study that showed decreased concentrations after betaine (Waldman et al., 2023). One study also measured carbohydrate and fat oxidation and found that after betaine supplementation, fat oxidation decreased, and carbohydrate utilization increased during graded exercise test (Waldman et al., 2023). Betaine concentration was measured in one study and showed a significant increase after supplementation and compared to placebo (Yang et al., 2020).

4. Discussion

This systematic review and meta-analysis aimed to comprehensively reassess the effects of betaine supplementation on physical performance considering recent studies published since the last systematic review on this topic (Ismaeel, 2017). The included studies employed various exercise performance tests, encompassing muscular strength, endurance, power, aerobic capacity, and specific functional tasks.

The analysis of maximal muscular strength outcomes indicated a significant overall effect of betaine supplementation, favouring betaine over placebo trials. This effect was primarily driven by improvements in lower body strength, including measures such as 1RM in SQ, LP, and sumo deadlift. However, the effect on upper body strength did not reach statistical significance, although a sensitivity analysis revealed potential benefits when certain studies were excluded. These findings suggest that betaine may have a more pronounced impact on lower body strength compared to upper body strength.

The observed improvement in lower body strength indicates that betaine may enhance the performance of large muscle groups, which are heavily involved in lower body compound exercises like SQ and deadlifts (Yang et al., 2022). It is important to note that the heterogeneity in the meta-analysis was moderate to high, indicating some variability in study results. This heterogeneity could be attributed to differences in study populations, training protocols, or dosages of betaine. However, the effect of betaine was not dependent on the duration of supplementation, age, sex, training status, or exercise type (for lower-body strength). We were unable to include betaine dose in the meta-regression because only one study used ahigher dose (6 g/day compared to the commonly used 2.0–2.5 g/day). Additionally, alimitation of the regression results is the low number of studies ($n < 10$) and the low number of studies including females (one study for both upper- and lower-body strength) or mixed samples (only one study for lower-body strength).

The mechanism by which betaine increases muscular strength in a relatively short period of time (several weeks) is still hypothetical and warrants further investigation. Betaine, together with 5-methyltetrahydrofolate, acts as a methyl donor for homocysteine methylation to methionine (Ueland, 2011). Betaine – homocysteine methyltransferase transfers a methyl group from betaine (Ganu et al., 2015). Subsequently, methionine is adenylated to S-adenosylmethionine, a universal methyl donor in over 50 transmethylation reactions (Schubert et al., 2003). S-adenosylmethionine expands body pools of transmethylation products, such as phosphatidylcholine and creatine. Betaine, as a methyl group donor in S-adenosylmethionine formation, plays a crucial role in the transmethylation process for creatine formation (El-Ghany Wa & Babazadeh, 2022). It has been reported that increasing the level of creatine in muscles depends on the capacity of betaine as a methyl donor (El-Ghany Wa & Babazadeh, 2022). Therefore, it seems reasonable to expect an increase in creatine availability with betaine supplementation (Zhan et al., 2006).

In the muscle cells creatine combines with phosphate to form phosphocreatine. Phosphocreatine buffers adenosine triphosphate levels to improve high-intensity exercise capacity, potentially allowing one to train with higher volumes during resistance training sessions (Kreider et al., 2017). One study directly measured the effect of betaine supplementation, either alone or in combination with creatine, on muscle creatine content. However, this study failed to demonstrate such a relationship. It is worth noting that the participants in the del Favero et al. (2012) study were untrained and were instructed to limit their training during the intervention. This limitation could have influenced the results, as mechanical tension is known to promote creatine uptake (Forbes & Candow, 2018).

The intensity of training may play a crucial role in the effectiveness of betaine supplementation, particularly in high-intensity strength or power routines that impose substantial metabolic stress. Under such conditions, the elevated cellular stress and metabolic demands may enhance betaine's role as an organic osmolyte. As an osmolyte, betaine stabilizes proteins, maintains cellular hydration, and protects sensitive metabolic pathways, including those involved in protein turnover, amino acid metabolism, pH regulation, and gene expression (Willingham et al., 2020). This protective effect is especially significant during high-intensity resistance exercise, where cellular stress can lead to enzyme inactivation, membrane instability, and increased expression of heat shock proteins (Willingham et al., 2020). Betaine's capacity to maintain cellular integrity and reduce fatigue may therefore contribute to improved performance and increased training volume, potentially enhancing strength gains. In fact, one study showed increased training voulme of BP and SQ at microcycle 3 (5th-6th week of betaine suppplementation) after betaine compared to placebo (Cholewa et al., 2013). Thus, the benefits of betaine might be more pronounced during high-intensity training, where its functions in maintaining biochemical balance and protecting metabolic pathways are most critical.

Betaine has demonstrated the ability to stimulate myotube differentiation and hypertrophy in laboratory settings, primarily through its impact on IGF-1 (R. Chen et al., 2022). Furthermore, in addition to its positive influence on muscle regeneration

(Senesi et al., 2013) betaine may also have anti-inflammatory properties, as evidenced by its capacity to inhibit the expression of adipokines induced by hypoxia (Olli et al., 2013). Furthermore, in a rodent model, betaine promoted muscle synthesis via tethering mTORC1 on the lysosomal membrane, activating mTORC1 signalling via increasing S-adenosylmethionine (S. Chen et al., 2021). Considering these findings, it is reasonable to infer that betaine might enhance athletic performance, diminish fatigue and enhance overall muscle function.

Despite the lack of a significant overall effect in the meta-analysis of muscular power outcomes, the potential improvement in VJ performance highlighted by the sensitivity analysis, excluding the study by Yang et al. (2022) due to its low quality and lack of randomization, suggests that betaine supplementation might still hold promise for enhancing explosive movements such as jumping. This potential improvement in VJ performance could be attributed to the aforementioned increased lower body strength resulting from betaine supplementation and possibly increased phosphocreatine synthesis. The correlation between countermovement jumps and lower body strength (1RM in half SQ) was previously reported (Boraczyński et al., 2020).

The analysis of muscular endurance outcomes did not reveal a significant effect of betaine supplementation. This result was consistent for both upper and lower body endurance measures. The lack of a substantial effect on muscular endurance suggests that betaine may primarily impact acute measures of strength rather than endurance performance. The diverse range of endurance tests employed in the included studies may contribute to the lack of a coherent effect.

Aerobic capacity outcomes were assessed in only a limited number of studies, not sufficient for meta-analysis calculations. While some studies reported no significant changes in parameters such as VO_2max and HR (Waldman et al., 2023; Yang et al., 2020) others observed an increase in estimated VO_2max following betaine supplementation (Nobari, Cholewa, et al., 2021). The limited number of studies and varying methodologies make it challenging to draw definitive conclusions regarding the impact of betaine on aerobic capacity.

Studies that evaluated conditioning tasks and CrossFit-related performance tests showed mixed results. We could not perform meta-analysis due to limited data. Betaine supplementation appeared to enhance performance in the Fight Gone Bad test (Zawieja et al., 2023). However, other tests, such as the Bergeron Beep Test and the 2-kilometre rowing test, did not show significant improvements (Moro et al., 2020).

Betaine supplementation was associated with improvements in body composition, including increased fat-free mass and reduced body fat in some studies (Cholewa et al., 2013, 2018; Waldman et al., 2023). In one study, lean body mass and arm cross-sectional area increased, but there were no significant improvements in muscular strength (squat or bench press) (Cholewa et al., 2013). Another study that reported increases in fat-free mass did not measure maximal strength, but observed increased total power in the third Wingate sprint (Waldman et al., 2023). However, the effects of betaine on body composition were not consistent across all studies (Moro et al., 2020; Zawieja et al., 2023). Despite animal research (Fu et al., 2023; Wang et al., 2021) and human observational studies (Chen et al., 2015) suggesting potential correlations between betaine and body fat, a recent meta-analysis revealed no such effect (Ashtary-Larky et al., 2022). Whether the observed increase in muscle strength after betaine supplementation is related to hypertrophy should be addressed further in future studies. There is limited data (only three studies) (Cholewa et al., 2013, 2018; Moro et al., 2020) on the parallel effects of betaine on lean body mass and muscular strength, making it difficult to draw any conclusions at this time.

Hormone concentrations, including IGF-1, testosterone, GH, and cortisol, exhibited variable responses to betaine supplementation. However, betaine appears to elevate IGF-1 and total testosterone levels, which may indirectly contribute to the observed effect of betaine on muscular strength (Arazi et al.,

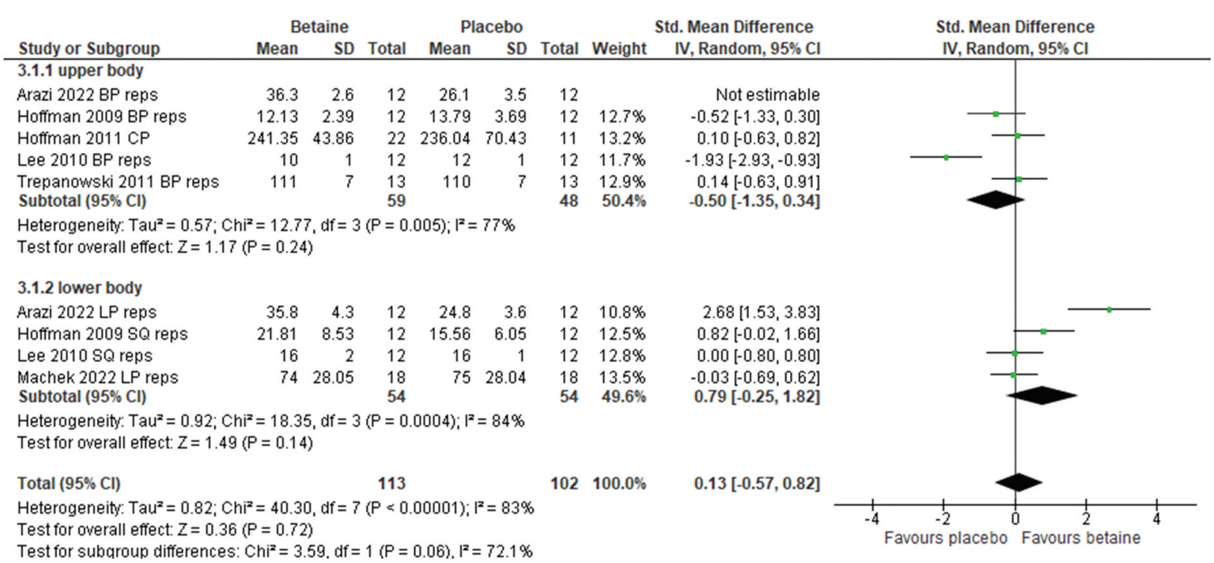

Figure 5. Meta-analysis results for muscular endurance. Abbreviation list: BP reps, repetitions to failure in bench press; CON, concentric endurance in chest press; ECC, eccentric endurance in chest press; HL, high load; LL, low load; LP reps, repetitions to failure in leg press; SQ reps, repetitions to failure in squat.

2022; Machek, Harris, et al., 2022; Nobari, Kargarfard, et al., 2021; Zawieja et al., 2023). Metabolic and biochemical parameters, such as lactate levels, muscle phosphocreatine, glucose, and lipid profiles, showed limited changes in response to betaine supplementation. It is plausible, though, that betaine does not affect energy production anaerobically from glucose. One study demonstrated that during graded exercise, betaine increased CHO oxidation and decreased fat oxidation, but without any alterations in lactate concentrations (Waldman et al., 2023). This suggests that betaine may influence fuel utilization during submaximal exercise. However, the available data is limited and should be corroborated by future studies.

Limitations and Future Directions

Several limitations of this meta-analysis should be acknowledged. The heterogeneity among studies was high for some outcomes (upper body strength, muscular endurance (total, upper and lower body), and BP throw). Two studies included in this review do not clearly state whether they were randomized (Yang et al., 2020, 2022). The meta-regression was limited by low number of studies for maximal strength outcomes ($n < 10$) and underrepresentation of female participants. Future studies should consider addressing gender differences in light of molecular data that indicate variations in the expression of genes encoding enzymes in one-carbon metabolism between males and females (Sadre-Marandi et al., 2018). For instance, betaine-homocysteine methyltransferase is downregulated in females (Sadre-Marandi et al., 2018). This enzyme plays a role in catalysing the methylation of homocysteine to methionine, with betaine serving as the methyl-group donor. Furthermore, the betaine content in the muscle tissue of female rats was nearly half that of male rats (Slow et al., 2009). Another limitation of this meta-analysis is that, in crossover studies, the short washout period – while sufficient to reduce betaine concentrations to baseline levels – may not be adequate to return performance to baseline levels. This particularly concerns participants who started with betaine, showed improved performance, and did not return to baseline during the washout period, thus potentially entering the placebo arm still benefiting from the adaptation effects caused by betaine (so called the carry-over effect). However, accounting for this factor in the meta-analysis was not possible, as the authors did not report data based on group allocation.

Practical applications

The present systematic review and meta-analysis suggest that chronic (minimum one week) betaine supplementation can be beneficial at doses ranging from 2.5 to 6 g per day for improving muscular strength, particularly in the lower body and possibly VJ performance. This information can be valuable for athletes and trainers who are considering incorporating betaine into their strength training programmes. Athletes who stand to benefit the most from betaine supplementation include weightlifters, powerlifters, bodybuilders, strongmen, football players, CrossFit practitioners, martial artists, and rugby players. To date, there is no evidence to support the ingestion of betaine peri-workout. It is recommended to divide the daily dose into smaller portions. Betaine can be administered in different forms, such as capsules or dissolved in water, with no evidence indicating the superiority of either method of administration (Atkinson et al., 2008).

5. Conclusions

In conclusion, this comprehensive meta-analysis indicates that chronic betaine supplementation may have a positive impact on total strength and lower body strength. Still, it does not consistently affect upper body strength, muscular power, and endurance. Betaine also has the potential to increase VJ performance. The effects of betaine on functional tasks, aerobic endurance and body composition are variable. The heterogeneity in study outcomes highlights the need for further research to elucidate the specific contexts in which betaine supplementation is most effective and to identify potential factors that influence individual responses.

Disclosure statement

No potential conflict of interest was reported by the author(s).

Funding

The author(s) reported there is no funding associated with the work featured in this article.

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Availability of data

Data extracted from included studies and data used for all analyses are available from the corresponding author on request.

Author contributions

EZ created the study protocol and search strategy; EZ and SM performed the search and data extraction; EZ and MW performed risk of bias assessment; EZ performed GRADE analysis and a statistical summary of the data; EZ, JC, and NEZ wrote the manuscript and interpreted the data.

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