British Journal of Nutrition (2018), **119**, 636–657
doi:10.1017/S0007114518000132

British Journal of Nutrition

Nitrate supplementation improves physical performance specifically in non-athletes during prolonged open-ended tests: a systematic review and meta-analysis

Helton O. Campos[1]†, Lucas R. Drummond[1]†, Quezia T. Rodrigues[1], Frederico S. M. Machado[1], Washington Pires[2], Samuel P. Wanner[3] and Cândido C. Coimbra[1]*

[1]*Departamento de Fisiologia e Biofísica, Instituto de Ciências Biológicas, Universidade Federal de Minas Gerais, Av. Antônio Carlos 6627, 31270-901, Belo Horizonte, Minas Gerais, Brazil*

[2]*Departamento de Educação Física, Universidade Federal de Juiz de Fora – Campus Governador Valadares, Av. Doutor Raimundo Monteiro Rezende 330, 35010-177, Governador Valadares, Minas Gerais, Brazil*

[3]*Departamento de Educação Física, Universidade Federal de Minas Gerais, Av. Antônio Carlos 6627, 31270-901, Belo Horizonte, Minas Gerais, Brazil*

(Submitted 31 March 2017 – Final revision received 6 December 2017 – Accepted 5 January 2018)

Abstract

Nitrate (NO_3^-) is an ergogenic nutritional supplement that is widely used to improve physical performance. However, the effectiveness of NO_3^- supplementation has not been systematically investigated in individuals with different physical fitness levels. The present study analysed whether different fitness levels (non-athletes *v.* athletes or classification of performance levels), duration of the test used to measure performance (short *v.* long duration) and the test protocol (time trials *v.* open-ended tests *v.* graded-exercise tests) influence the effects of NO_3^- supplementation on performance. This systematic review and meta-analysis was conducted and reported according to the guidelines outlined in the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) statement. A systematic search of electronic databases, including PubMed, Web of Science, SPORTDiscus and ProQuest, was performed in August 2017. On the basis of the search and inclusion criteria, fifty-four and fifty-three placebo-controlled studies evaluating the effects of NO_3^- supplementation on performance in humans were included in the systematic review and meta-analysis, respectively. NO_3^- supplementation was ergogenic in non-athletes (mean effect size (ES) 0·25; 95 % CI 0·11, 0·38), particularly in evaluations of performance using long-duration open-ended tests (ES 0·47; 95 % CI 0·23, 0·71). In contrast, NO_3^- supplementation did not enhance the performance of athletes (ES 0·04; 95 % CI −0·05, 0·15). After objectively classifying the participants into different performance levels, the frequency of trials showing ergogenic effects in individuals classified at lower levels was higher than that in individuals classified at higher levels. Thus, the present study indicates that dietary NO_3^- supplementation improves physical performance in non-athletes, particularly during long-duration open-ended tests.

Key words: Diet: Fitness level: Nitric oxide: Fatigue

Nitrate (NO_3^-) is an ergogenic nutritional supplement widely consumed by exercise practitioners and athletes to improve their health and physical performance[1]. The widespread use of NO_3^- likely reflects its abundant availability in many vegetables, and its content ranges from <20 mg/100 g in sweet potato to >250 mg/100 g in beetroot[2]. Although oral bacteria can reduce NO_3^- to nitrite (NO_2^-), the transit of these foods in the mouth is short, and the resulting increase in NO_3^- bioavailability appears to be related to the intrinsic NO_3^- content in the vegetable or supplement. Indeed, increased NO_3^- bioavailability could favour nitric oxide (NO) synthesis[3]. NO is a signalling molecule associated with improved cardiovascular and skeletal muscle functions that may potentially enhance physical performance and even facilitate adaptations to exercise training[4]. Nevertheless, the scientific literature provides controversial results regarding the performance-enhancing effects induced by NO_3^- supplementation.

Two systematic reviews and meta-analyses on this topic have recently been published, establishing clear practical recommendations and directions for future studies investigating changes in performance induced by NO_3^- supplementation[5,6]. Hoon *et al.*[5] and McMahon *et al.*[6] analysed data according to the exercise protocol used (i.e. time trials, open-ended tests and graded-exercise tests) and observed that dietary NO_3^- supplementation improved endurance only when performance was evaluated using open-ended tests. Notably, none of these two

Abbreviation: PL, performance level.

* **Corresponding author:** C. C. Coimbra, fax +55 31 3409 2924, email coimbrac@icb.ufmg.br

† These authors contributed equally to this work.

meta-analyses divided and analysed separately the studies conducted with athletes or non-athletes, as we are proposing here. McMahon *et al*. performed a continuous variable meta-regression analysis and reported that the fitness level did not have an influence on the ergogenic effect of dietary NO_3^- supplementation[6]. However, grouping the data according to the exercise protocol may result in an important bias. In fact, the studies using open-ended tests were mainly performed in non-athletes. In contrast, most studies using time trials were performed in athletes. This disparity might have led to a misinterpretation of the results owing to an unintentional division based on individuals' physical fitness level. Interestingly, neither of the two recent systematic reviews addressed the following question raised by Jonvik *et al*.: 'Can elite athletes benefit from dietary nitrate supplementation?'[7–9]. Therefore, information regarding the effectiveness of NO_3^- supplementation in individuals with different physical fitness levels is lacking. Moreover, physical performance is modulated by various mechanisms and depends on several factors, including the duration of the test performed (i.e. short or long duration). Thus, the influence of the test duration on the changes in performance induced by NO_3^- supplementation in individuals with different fitness levels remains to be investigated.

Increased NO availability resulting from NO_3^- supplementation has beneficial effects on health and physical performance and has been largely studied in humans and laboratory animals. In the central nervous system, NO prevented exaggerated increases in the core body temperature in rats subjected to exercise by increasing cutaneous heat loss and decreasing the metabolic cost of running[10–13]. In these rat studies, the pharmacological blockade of central NO synthesis markedly impaired endurance[10,12], whereas an increased NO availability in the brain did not affect endurance[13]. In humans, the physical performance benefits mediated by dietary NO_3^- supplementation have been attributed to peripheral effects, including reduced arterial pressure and VO_2. The latter effect leads to a reduced oxygen cost during exercise that is most likely due to the reduced cost of ATP for muscle force production, improved mitochondrial efficiency and increased muscle oxygenation[14,15]. In contrast, the adverse events related to NO_3^- supplementation are minor and restricted to red urine (beeturia) and stool, which usually results from the ingestion of beetroot in juice or meals[16,17].

Interestingly, both acute and chronic supplementations of NO_3^- have been shown to either improve[18–24] or have no effect[14,25–29] on endurance performance. The uncertain efficacy of NO_3^- supplementation appears to be related to the fitness level of the investigated population as demonstrated by a careful evaluation of the cumulative number of trials reporting the performance benefits or lack thereof in both non-athletes (healthy individuals engaged in regular physical activity but not involved in sports competitions) and athletes (Fig. 1). Notably, nearly 65 % of the publications on this topic did not report the benefits resulting from NO_3^- supplementation. However, if only those studies performed in non-athletes are considered, approximately 45 % of the publications show a supplementation-mediated positive effect on physical performance, whereas the percentage of papers showing beneficial effects in athletes is lower than 30 % (Fig. 1). Collectively, these observations reinforce the relevance of the present systematic review and meta-analysis, which help clarify the contradictory reports of the effects of NO_3^- supplementation on physical performance.

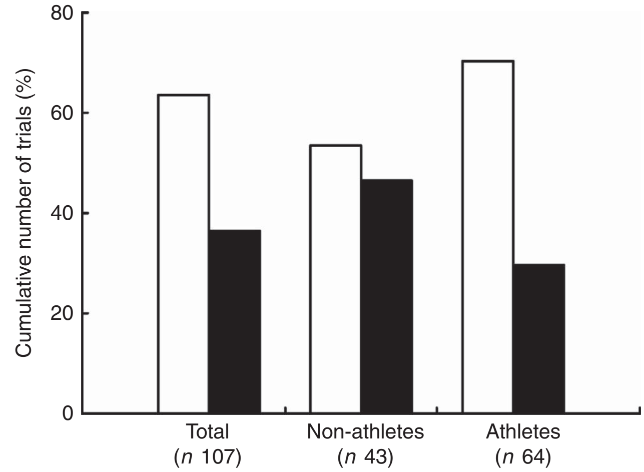

Fig. 1. Number of trials (%) reporting that dietary NO_3^- supplementation had no effect (□) and/or a positive effect (■) on physical performance in non-athletes and athletes.

Therefore, the present study systematically analysed whether different physical fitness levels (i.e. non-athletes *v.* athletes) influence the effects of NO_3^- supplementation on physical performance. In addition, we also evaluated the influence of the duration of the tests used to measure performance (i.e. short *v.* long duration) and the test protocol used (i.e. time trials *v.* open-ended tests *v.* graded-exercise tests) on the effect of NO_3^- supplementation on physical performance in individuals with different physical fitness levels. Thus, the present analyses provide information that is useful to exercise practitioners, athletes, coaches and conditioning professionals who are interested in improving physical performance and achieving health benefits.

Methods

Search strategy

This systematic review and meta-analysis was conducted and reported according to the guidelines outlined in the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) statement[30,31]. A systematic search of electronic databases, including PubMed, Web of Science, SPORTDiscus and ProQuest, was performed in August 2017 without any date restrictions. The search strategy was supplemented by manual cross-matching of each publication reference list and key author searches. Combinations of the following keywords were used: effort, endurance, exercise, fatigue, nitrate, nitrate supplementation, nitrite, nitrite supplementation, performance, power, running, speed, sport and workload.

Study selection

Studies that met the following criteria were included in this systematic review and meta-analysis: (i) the participants were healthy humans (either non-athletes or athletes), (ii) physical performance was measured after the participants were supplemented with NO_3^- and (iii) the studies were placebo-controlled trials. Furthermore, all included studies were written in English. Reviews, summaries, case studies and letters were not included, although this bibliography was consulted. Studies involving hypoxic conditions, individuals with diseases, exercise in the heat, children and elderly people, and laboratory animals were excluded. On the basis of the search and inclusion/exclusion criteria, fifty-four studies (106 trials) were selected for inclusion in this systematic review, and fifty-three studies (104 trials) were included in the meta-analysis (Fig. 2). Notably, several studies measured more than one physical performance parameter. The data addressing the effect of NO_3^- supplementation on each parameter were included, and therefore the number of trials was greater than the number of studies. Only one study with one trial[20] and one trial in a study with several trials[32] were excluded from the meta-analysis because they did not include the standard deviation data needed to calculate the effect size.

Data grouping

The selected studies were divided into the following two groups according to the physical fitness level of the individuals tested: non-athletes (forty-three trials) and athletes (sixty-three trials). The individuals were allocated into these two groups according to the classification used by the authors of the research papers, which were consulted. This strategy was efficient in dividing the participants into two groups with different functional capacities as demonstrated by the higher VO_{2max} values in the athletes than in the non-athletes (61·1 (SD 1·8) *v.* 50·5 (SD 1·8) ml/kg per min; *t*-test, $P < 0·05$). Similarly, the studies selected for inclusion in the meta-analysis were initially divided into the following two groups: non-athletes (forty-three trials) and athletes (sixty-one trials). The two groups were then subdivided according to the duration of the test performed as follows: short duration (non-athletes, eighteen trials; athletes, seventeen trials) or long duration (non-athletes, twenty-five trials; athletes, forty-four trials). Exercises lasting less than 180 s, thereby characterised by a relevant anaerobic contribution to the energy expenditure, were considered short-duration exercises. Alternatively, exercise bouts lasting more than 180 s were considered long-duration exercises[33]. In addition, because NO_3^- supplementation has been shown to have a positive effect on physical performance only in non-athletes during long-duration tests, this group was further subdivided according to the test protocol used (open-ended tests (constant power), fourteen trials; time trials, four trials; and graded-exercise tests (incremental power), five trials). Open-ended tests consist of exercising at a constant power until the participant is volitionally fatigued; the time until fatigue, which may be highly variable among subjects, is considered the main measure of performance in this test. Finally, owing to the large number of studies in cycling athletes, a specific analysis was conducted for this sport (thirty-seven trials).

Analysis of the relationship between the performance level and the response to NO_3^- supplementation

Because the authors of the research papers may have been imprecise in the classification of their subjects as athletes, we decided to perform an objective analysis. Thus, the individuals were grouped into different performance levels (PL) according to the classification provided by De Pauw *et al.*[34]. These authors divided the participants in sport science studies into the following five different levels: performance level 1 (PL1) included untrained and sedentary subjects with a $VO_{2max} < 45·0$ ml/kg per min; performance level 2 (PL2) included recreationally trained subjects with a VO_{2max} between 45·0 and 54·9 ml/kg per min; performance level 3 (PL3) included trained subjects with a VO_{2max} between 55·0 and 64·9 ml/kg per min; performance level 4 (PL4) included highly trained subjects with a VO_{2max} between 65·0 and 71·0 ml/kg per min; and performance level 5 (PL5) included professional subjects with a $VO_{2max} > 71·0$ ml/kg per min. On the basis of this study, we grouped the individuals into five levels and then evaluated the relationship between the PL and the changes in performance induced by NO_3^- supplementation.

Risk of bias assessment

Two independent reviewers assessed the risk of bias using an adapted Grading of Recommendations Assessment, Development and Evaluation (GRADE) instrument[35]. Discrepant evaluations were settled via discussion with a third reviewer. Using this approach, it was possible to evaluate the risk of bias in each study included in the present systematic review. Domains reflecting sequence generation, allocation concealment, blinding of participants and personnel, incomplete outcome data, selective outcome reporting and other sources of bias were evaluated.

Statistical analysis

The mean and standard deviation values of the performance indexes in both the NO_3^- supplementation and control trials were obtained from the data provided in the consulted research papers. Heterogeneity was evaluated using the χ^2 test for homogeneity and the I^2 statistic. The effect size (Cohen's *d* or Hedges' *g*) was calculated for the performance indexes in each study. Then, a weighted-mean estimate of the effect size was calculated to account for differences in the sample sizes. The mean unweighted effect size and associated 95 % CI were also calculated. We used Cohen's classification of the effect size magnitude, where $d < 0·20$ for negligible effect; $d = 0·20–0·49$ for small effect; $d = 0·50–0·79$ for moderate effect; and $d > 0·8$ for large effect[36]. The χ^2 test was used to compare the frequency of trials showing improved performance in response to NO_3^- supplementation among the different PL. Student's *t* test was used to compare the VO_{2max} between the non-athletes and athletes. Pearson's correlations were performed to evaluate the association between the supplementation parameters (dose, number of days and total amount ingested) and the changes in physical performance. Publication bias was assessed by a visual inspection of funnel plots of the standard error *v.* effect size[37].

Results

Systematic review

In total, 4732 studies were identified through the database and reference searches. After removing the duplicates and

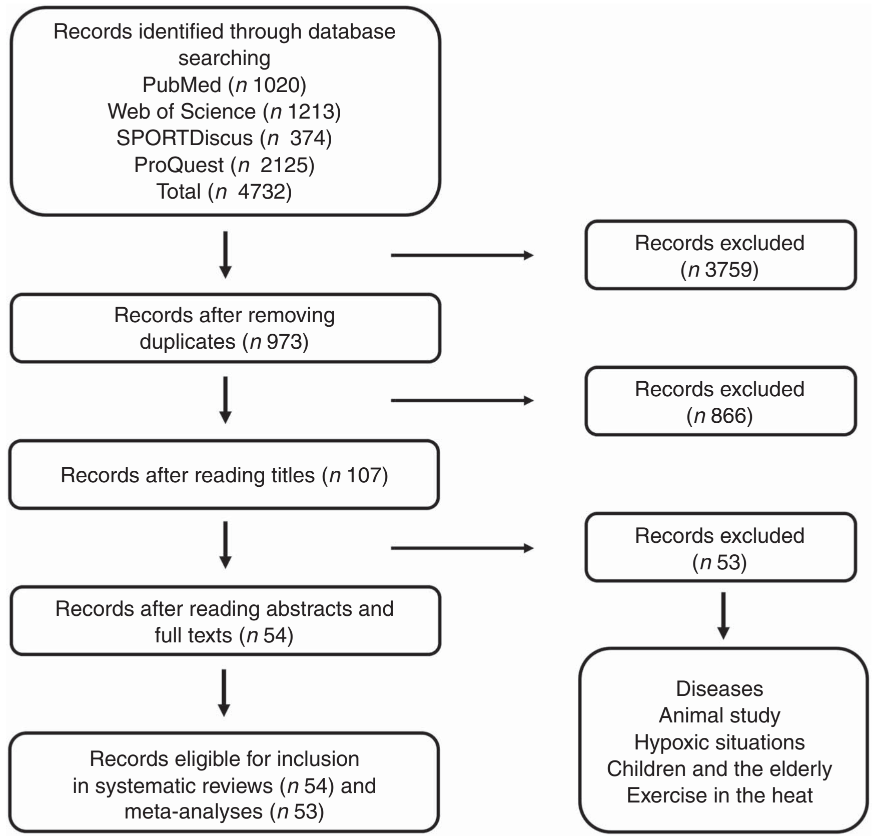

Fig. 2. Summary of the study selection process.

excluding papers that did not meet the eligibility criteria according to a review of their titles, abstracts and full texts, fifty-four studies (106 trials and 662 individuals) were selected for inclusion in the systematic review (Fig. 2).

The characteristics of the subjects, including information regarding the supplementation regimens and effects of NO_3^- supplementation on the physical performance of non-athletes and athletes in each study, are summarised in Tables 1 and 2, respectively. Notably, most studies used beetroot juice as a form of NO_3^- supplementation. However, these studies were heterogeneous in several supplementation features, including the ingested volume (70, 140, 250, 280 or 500 ml), dose (between 4·0 and 19·5 mmol), days of supplementation (between 1 and 15 d), timing of supplementation before the trial (between 40 and 1440 min) and the parameter measured to determine physical performance.

Meta-analyses

In total, fifty-three studies (104 trials and 648 individuals) were included in the meta-analysis.

Non-athletes. After pooling the data from forty-three trials, the mean effect size was 0·25 (95 % CI 0·11, 0·38), which indicates that the dietary NO_3^- supplementation had a small and significant beneficial effect on physical performance ($P < 0·05$; Fig. 3). According to a fixed-effects analysis, no heterogeneity was observed among these studies ($I^2 = 0$ %; $Q = 15·26$, df = 42, $P = 1·00$).

Athletes. After pooling the data from sixty-one trials, the mean effect size was 0·04 (95 % CI –0·05, 0·15), which indicates that the dietary NO_3^- supplementation had a negligible and non-significant effect on improving physical performance ($P > 0·05$; Fig. 4). According to a fixed-effects analysis, no heterogeneity was observed among these studies ($I^2 = 0$ %; $Q = 18·16$, df = 60, $P = 1·00$). The subsequent analysis consisted of subdividing both the athletes and non-athletes into those performing short- and long-duration tests.

Non-athletes subjected to short-duration tests. After pooling the data from eighteen trials, the mean effect size was 0·12 (95 % CI –0·07, 0·31), which indicates that the dietary NO_3^- supplementation had a negligible and non-significant effect on physical performance ($P > 0·05$; Fig. 5). According to a fixed-effects analysis, no heterogeneity was observed among these studies ($I^2 = 0$ %; $Q = 4·43$, df = 17, $P = 0·99$).

Athletes subjected to short-duration tests. After pooling the data from seventeen trials, the mean effect size was 0·03 (95 % CI –0·17, 0·23), which indicates that the dietary NO_3^- supplementation had a negligible and non-significant effect on performance ($P > 0·05$; Fig. 6). According to a random-effects

https://doi.org/10.1017/S0007114518000132 Published online by Cambridge University Press

Table 1. Study characteristics – non-athletes
(Mean values and standard deviations)

			Nitrate supplementation									
			VO_{2peak}/VO_{2max} (ml/kg per min)									
References	No. of subjects (♂, ♀)	Characteristics of subjects	Mean	SD	Ingested fluid/ volume (ml)	Dose (mmol)	Placebo substance	Days of supplementation	Time before trial (min)	Exercise protocol	Variable of physical performance	Results
Aucouturier *et al.* (1)[38]	12 (♂)	Healthy	46·6	3·4	BJ/500	5·4	Apple blackcurrant juice	3	90	Supramaximal intermittent exercise test	Work (kJ)	S = 168·1 (SD 60·2) NS = 142·0 (SD 46·8) D = no
Aucouturier *et al.* (2)[38]	12 (♂)	Healthy	46·6	3·4	BJ/500	5·4	Apple blackcurrant juice	3	90	Supramaximal intermittent exercise test	Time (min)	S = 19·6 (SD 8·1) NS = 16·4 (SD 6·0) D = yes
Bailey *et al.*[18]	8 (♂)	Healthy	49	5	BJ/500	5·5	Blackcurrant cordial without nitrate	6	NR	Severe-intensity exercise	Time (s)	S = 675 (SD 203) NS = 583 (SD 145) D = yes
Bailey *et al.* [19]	7 (♂)	Healthy, recreationally active	–		BJ/500	5·1	Low-energy blackcurrant juice cordial	6	NR	High-intensity exercise	Time (s)	S = 734 (SD 288) NS = 586 (SD 211) D = no
Bailey *et al.* (1)[39]	7 (♂)	Healthy	–		BJ/70	6·2	Sodium chloride	9	150	Cycling at 35 rpm	Time (s)	S = 344 (SD 74) NS = 341 (SD 99) D = no
Bailey *et al.* (2)[39]	7 (♂)	Healthy	–		BJ/70	6·2	Sodium chloride	9	150	Cycling at 115 rpm	Time (s)	S = 362 (SD 137) NS = 297 (SD 79) D = yes
Breese *et al.*[40]	9 (4 ♂ and 5 ♀)	Healthy, physically active	♂ = 3·73 ♀ = 2·69	♂ = 0·46* ♀ = 0·52*	BJ/140	8·0	BJ negligible nitrate content	6	120	Step exercise tests until fatigue	Time (s)	S = 635 (SD 258) NS = 521 (SD 158) D = yes
Buck *et al.*[41]	13 (♀)	Amateur team-sport participants	–		BJ/70	6·0	BJ negligible nitrate content	1	180	3 sessions of 6 × 20 m sprints	Total sprint time (s)	S = 69·8 (SD 4·9) NS = 69·9 (SD 4·1) D = no
Christensen *et al.* (1)[42]	8 (♂)	Recreationally active	46	3	BJ/150	9	Blackcurrant citrus with 0·2 mmol nitrate	1	180–249	Incremental leg exercise	Peak power output (W)	S = 304 (SD 34) NS = 310 (SD 47) D = no
Christensen *et al.* (2)[42]	8 (♂)	Recreationally active	46	3	BJ/150	9	Blackcurrant citrus with 0·2 mmol nitrate	1	180–249	Incremental arm exercise	Peak power output (W)	S = 121 (SD 13) NS = 117 (SD 14) D = no
Coggan *et al.* (1)[43]	12 (7 ♂ and 5 ♀)	Healthy	–		BJ/140	11·2	BJ negligible nitrate content	1	120	Knee extensor contractile function (1·57 rad/s)	Peak power output (W/kg)	S = 3·31 (SD 0·55) NS = 3·38 (SD 0·72) D = no
Coggan *et al.* (2)[43]	12 (7 ♂ and 5 ♀)	Healthy	–		BJ/140	11·2	BJ negligible nitrate content	1	120	Knee extensor contractile function (3·14 rad/s)	Peak power output (W/kg)	S = 5·38 (SD 1·10) NS = 5·48 (SD 1·31) D = no
Coggan *et al.* (3)[43]	12 (7 ♂ and 5 ♀)	Healthy	–		BJ/140	11·2	BJ negligible nitrate content	1	120	Knee extensor contractile function (4·17 rad/s)	Peak power output (W/kg)	S = 6·76 (SD 1·59) NS = 6·67 (SD 1·73) D = no
Coggan *et al.* (4)[43]	12 (7 ♂ and 5 ♀)	Healthy	–		BJ/140	11·2	BJ negligible nitrate content	1	120	Knee extensor contractile function (6·28 rad/s)	Peak power output (W/kg)	S = 7·64 (SD 1·80) NS = 7·34 (SD 1·87) D = yes
Corry *et al.*[44]	10 (♂)	Recreationally active	–		BJ/140	8·0	Low-energy blackcurrant juice with negligible NO_3^-	2	40	Wingate test	Mean power output (W/kg)	S = 7·95 (SD 0·55) NS = 7·63 (SD 0·91) D = no
Fulford *et al.* (1)[45]	8 (♂)	Healthy, physically active	–		BJ/250	10·2	BJ negligible nitrate content	1	150	Isometric maximum voluntary contraction protocol	Mean force of peak contraction (N)	S = 368 (SD 90) NS = 382 (SD 143) D = no
Fulford *et al.* (2)[45]	8 (♂)	Healthy, physically active	–		BJ/250	10·2	BJ negligible nitrate content	5 (2×/d)	150	Isometric maximum voluntary contraction protocol	Mean force of peak contraction (N)	S = 380 (SD 65) NS = 387 (SD 119) D = no
Fulford *et al.* (3)[45]	8 (♂)	Healthy, physically active	–		BJ/250	10·2	BJ negligible nitrate content	15 (2×/d)	150	Isometric maximum voluntary contraction protocol	Mean force of peak contraction (N)	S = 408 (SD 110) NS = 365 (SD 115) D = no

Table 1. *Continued*

			Nitrate supplementation									
			VO_{2peak}/VO_{2max} (ml/kg per min)									
References	No. of subjects (♂, ♀)	Characteristics of subjects	Mean	SD	Ingested fluid/ volume (ml)	Dose (mmol)	Placebo substance	Days of supplementation	Time before trial (min)	Exercise protocol	Variable of physical performance	Results
Kelly *et al.* (1)[46]	9 (♂)	Recreationally active	54·5	7·5	BJ/500 (250 + 250)	8·2	BJ negligible nitrate content	7–12	150	Severe-intensity exercise (60 % peak power output)	Time (s)	S = 696 (SD 120) NS = 593 (SD 68) D = yes
Kelly *et al.* (2)[46]	9 (♂)	Recreationally active	54·5	7·5	BJ/500 (250 + 250)	8·2	BJ negligible nitrate content	7–12	150	Severe-intensity exercise (70 % peak power output)	Time (s)	S = 452 (SD 106) NS = 390 (SD 86) D = yes
Kelly *et al.* (3)[46]	9 (♂)	Recreationally active	54·5	7·5	BJ/500 (250 + 250)	8·2	BJ negligible nitrate content	7–12	150	Severe-intensity exercise (80 % peak power output)	Time (s)	S = 294 (SD 50) NS = 263 (SD 50) D = yes
Kelly *et al.* (4)[46]	9 (♂)	Recreationally active	54·5	7·5	BJ/500 (250 + 250)	8·2	BJ negligible nitrate content	7–12	150	Severe-intensity exercise (100 % peak power output)	Time (s)	S = 182 (SD 37) NS = 166 (SD 26) D = no
Kokkinoplitis and Chester[47]	7 (♂)	Healthy	–		BJ/70	6·4	Blackcurrant juice	1	180	Repeated high-intensity sprints (5 × 6 s)	Mean peak power output (W)	S = 4133·5 (SD 674·4) NS = 3938·3 (SD 603·1) D = no
Lansley *et al.*[23]	9 (♂)	Physically active	55	7	BJ/500	6·2	BJ negligible nitrate content	6	180	Severe-intensity running	Time (min)	S = 8·7 (SD 1·8) NS = 7·6 (SD 1·5) D = yes
Larsen *et al.*[15]	9 (7 ♂ and 2 ♀)	Healthy	3·72	0·33*	Sodium nitrate	0·033 mmol/kg body mass	Sodium chloride	2 (3 × /d)	40	Incremental exercise on ergometers	Time (s)	S = 563 (SD 90) NS = 524 (SD 93) D = no
Mosher *et al.*[48]	12 (♂)	Recreationally active	–		BJ/70	6·4	Blackcurrant placebo drink	6	NR	Bench press exercise 3 sets until failure – 60 % 1RM	Total weight lifted (kg)	S = 2582·8 (SD 863·9) NS = 2171·5 (SD 720·5) D = yes
Murphy *et al.*[49]	11 (5 ♂ and 6 ♀)	Recreationally fit	–		Baked beetroot	8·0	Cranberry relish	1	60	Time trial 5 km	Running speed (km/h)	S = 12·3 (SD 9·0) NS = 11·9 (SD 8·6) D = no
Nyakayiru *et al.*[50]	32 (♂)	Soccer players	–		BJ/140	12·9	BJ negligible nitrate content	6	240	Yo-Yo test	Distance (m)	S = 1623 (SD 48) NS = 1574 (SD 47) D = yes
Porcelli *et al.* (1)[51]	8 (♂)	Healthy individuals with a low aerobic capacity	28·2–44·1		Sodium nitrate	5·5	Sodium chloride	6	210	Time trial 3 km	Time (s)	S = 886 (SD 74) NS = 910 (SD 82) D = yes
Porcelli *et al.* (2)[51]	7 (♂)	Healthy individuals with a moderate aerobic capacity	45·5–57·1		Sodium nitrate	5·5	Sodium chloride	6	210	Time trial 3 km	Time (s)	S = 723 (SD 90) NS = 734 (SD 93) D = yes
Porcelli *et al.* (3)[51]	6 (♂)	Healthy individuals with a high aerobic capacity	63·9–81·7		Sodium nitrate	5·5	Sodium chloride	6	210	Time trial 3 km	Time (s)	S = 627 (SD 30) NS = 629 (SD 28) D = no
Rienks *et al.*[52]	10 (♀)	Healthy	37·1	5·3	BJ/140	12·9	BJ negligible nitrate content	1	150	20 min of cycling exercise at RPE 13	Total mechanical work (kJ)	S = 30·3 (SD 5·3) NS = 29·8 (SD 6·1) D = no
Thompson *et al.*[53]	16 (♂)	Healthy, recreationally active	47·3	6·3	BJ/500	5·0	BJ negligible nitrate content	1	90	Continuous cycle exercise test until volitional exhaustion	Exercise tolerance (s)	S = 185 (SD 122) NS = 160 (SD 109) D = yes
Thompson *et al.*[54]	16 (♂)	Recreational team-sport players	50	7	BJ/70	6·4	BJ negligible nitrate content	7 (2 × /d)	150	Intermittent-sprint test	Total work done during the sprints (kJ)	S = 123 (SD 19) NS = 119 (SD 17) D = yes
Vanhatalo *et al.*[16]	8 (5 ♂ and 3 ♀)	Healthy	–		BJ/500	5·2	Low-energy blackcurrant juice cordial with low nitrate	15 (2 × /d)	150–180	Incremental cycling test	Peak power output (W)	S = 323 (SD 68) NS = 331 (SD 68) D = yes
Vasconcellos *et al.*[55]	25 (14 ♂ and 11 ♀)	Healthy	♂ = 64·31 ♀ = 52·79	♂ = 4·71 ♀ = 4·57	Two beetroot gels with 50 g each and 300 ml of water	9·92 (SD 1·97)	Placebo gel	1	90	Severe-intensity running	Time (s)	S = 395·4 (SD 179·6) NS = 390·9 (SD 158·5) D = no

https://doi.org/10.1017/S0007114518000132 Published online by Cambridge University Press

Table 1. *Continued*

References	No. of subjects (♂, ♀)	Characteristics of subjects	Nitrate supplementation: VO_{2peak}/VO_{2max} (ml/kg per min) Mean	SD	Ingested fluid/ volume (ml)	Dose (mmol)	Placebo substance	Days of supplementation	Time before trial (min)	Exercise protocol	Variable of physical performance	Results
Wylie *et al.* (1)[17]	10 (♂)	Healthy, recreationally active	–		BJ/70	4·2	Water	1	150	Severe-intensity cycling exercise	Time (s)	S = 508 (SD 102) NS = 470 (SD 81) D = no
Wylie *et al.* (2)[17]	10 (♂)	Healthy, recreationally active	–		BJ/140	8·4	Water	1	150	Severe-intensity cycling exercise	Time (s)	S = 570 (SD 153) NS = 498 (SD 113) D = yes
Wylie *et al.* (3)[17]	10 (♂)	Healthy, recreationally active	–		BJ/280	12·8	Water	1	150	Severe-intensity cycling exercise	Time (s)	S = 552 (SD 117) NS = 493 (SD 114) D = yes
Wylie *et al.*[56]	14 (♂)	Recreational team-sport players	52	7	BJ/140	4·1	BJ negligible nitrate content	2	150	Yo-Yo IR1	Distance covered (m)	S = 1704 (SD 304) NS = 1636 (SD 288) D = yes
Wylie *et al.* (1)[57]	10 (♂)	Recreational team-sport players	58	8	BJ/140	8·2	BJ negligible nitrate content	3	150	Maximal efforts (24 × 6-s protocol)	Mean power output (W)	S = 568 (SD 136) NS = 539 (SD 136) D = yes
Wylie *et al.* (2)[57]	10 (♂)	Recreational team-sport players	58	8	BJ/140	8·2	BJ negligible nitrate content	4	150	Maximal efforts (7 × 30-s protocol)	Mean power output (W)	S = 558 (SD 95) NS = 562 (SD 94) D = no
Wylie *et al.* (3)[57]	10 (♂)	Recreational team-sport players	58	8	BJ/140	8·2	BJ negligible nitrate content	5	150	Maximal efforts (6 × 60-s protocol)	Mean power output (W)	S = 374 (SD 57) NS = 375 (SD 59) D = no

♂, Male; ♀, female; BJ, beetroot juice; NR, not reported; S, supplemented; NS, no supplementation; D, statistical difference.
* Absolute VO_2 data in l/min.

https://doi.org/10.1017/S0007114518000132 Published online by Cambridge University Press

Table 2. Study characteristics – athletes
(Mean values and standard deviations)

			Nitrate supplementation									
			VO_{2peak}/VO_{2max} (ml/kg per min)									
References	No. of subjects (♂, ♀)	Characteristics of subjects	Mean	SD	Ingested fluid/ volume (ml)	Dose (mmol)	Placebo substance	Days of supplementation	Time before trial (min)	Exercise protocol	Measure of physical performance	Results
Bescós *et al.*[26]	11 (♂)	Cyclists and triathletes	65·1	6·2	Sodium nitrate/250	11·8	Sodium chloride	1	180	Incremental exercise	Time (s)	S = 416 (SD 32) NS = 409 (SD 27) D = no
Bescós *et al.* (1)[25]	13 (♂)	Cyclists and triathletes	–		Sodium nitrate/250	11·6	Sodium chloride	3	180	Distance trial (40 min) in cycle ergometer	Distance (km)	S = 26·4 (SD 1·1) NS = 26·3 (SD 1·2) D = no
Bescós *et al.* (2)[25]	13 (♂)	Cyclists and triathletes	–		Sodium nitrate/250	11·6	Sodium chloride	3	180	Distance trial (40 min) in cycle ergometer	Mean power output (W)	S = 258 (SD 28) NS = 257·3 (SD 28) D = no
Bond *et al.*[20]	14 (♂)	Rowers	–		BJ/500 (250 + 250)	5·0	Blackcurrant juice	6	NR	6 × 500 m rowing – ergometer repetitions at maximal intensity	Time (s)	S = 89·4 NS = 90·1 D = no
Boorsma *et al.* (1)[58]	8 (♂)	Distance runners	80	5	BJ/210 (on the test day) and 140 (other days)	19·5	BJ negligible nitrate content	1	150	Time trial 1500 m	Time (s)	S = 250·7 (SD 4·3) NS = 250·4 (SD 7·0) D = no
Boorsma *et al.* (2)[58]	8 (♂)	Distance runners	80	5	BJ/210 (on the test day) and 140 (other days)	19·5 (on the test day) and 13 (other days)	BJ negligible nitrate content	8	150	Time trial 1500 m	Time (s)	S = 250·5 (SD 6·2) NS = 251·4 (SD 7·6) D = no
Callahan *et al.* (1)[59]	8 (♂)	Endurance-trained cyclists	65·2	4·2	Gelatine capsules + water (400 ml)	5·0	Gelatine capsules (90 % BeetEssence and 10 % Black Cherry cool-aid)	3	60	Time trial 4000 m	Mean power output (W)	S = 388 (SD 54) NS = 386 (SD 56) D = no
Callahan *et al.* (2)[59]	8 (♂)	Endurance-trained cyclists	65·2	4·2	Gelatine capsules + water (400 ml)	5·0	Gelatine capsules (90 % Beet Essence and 10 % Black Cherry cool-aid)	3	60	Time trial 4000 m	Time (s)	S = 337·4 (SD 17·1) NS = 338·1 (SD 18·0) D = no
Cermak *et al.* (1)[21]	12 (♂)	Cyclists and triathletes	58	2	BJ/140 (70 + 70)	8·0	BJ negligible nitrate content	6	150	Time trial 10 km	Time (s)	S = 953 (SD 72·5) NS = 965 (SD 72·5) D = yes
Cermak *et al.* (2)[21]	12 (♂)	Cyclists and triathletes	58	2	BJ/140 (70 + 70)	8·0	BJ negligible nitrate content	6	150	Time trial 10 km	Mean power output (W)	S = 294 (SD 41·5) NS = 288 (SD 41·5) D = yes
Cermak *et al.* (1)[27]	20 (♂)	Cyclists or triathletes	60	1	BJ/140	8·7	BJ negligible nitrate content	1	150	Time trial approximately 1073 kJ	Time (min)	S = 65·5 (SD 4·8) NS = 65·0 (SD 4·8) D = no
Cermak *et al.* (2)[27]	20 (♂)	Cyclists or triathletes	60	1	BJ/140	8·7	BJ negligible nitrate content	1	150	Time trial approximately 1073 kJ	Mean power output (W)	S = 275 (SD 30·9) NS = 278 (SD 30·9) D = no
Christensen *et al.* (1)[32]	10 (♂)	Cyclists	72·1	4·5	BJ/500	8·0	Apple and blackcurrant juice	4	180	Repeated sprint test (6, 20 s)	Mean power output (W)	S = 630 (SD 84) NS = 630 (SD 92) D = no
Christensen *et al.* (2)[32]	10 (♂)	Cyclists	72·1	4·5	BJ/500	8·0	Apple and blackcurrant juice	6	180	Time trial 1677 kJ (400 kcal)	Time (min)	S = 18·33 NS = 18·61 D = no
Christensen *et al.* (3)[32]	10 (♂)	Cyclists	72·1	4·5	BJ/500	8·0	Apple and blackcurrant juice	6	180	Time trial 1677 kJ (400 kcal)	Mean power output (W)	S = 290 (SD 43 NS = 285 (SD 44) D = no
Christensen *et al.* (3)[42]	9 (♂)	Endurance-trained cyclists	64	3	BJ/150	9	Blackcurrant citrus with 0·2 mmol nitrate	1	180–249	Incremental leg exercise	Peak power output (W)	S = 418 (SD 47) NS = 406 (SD 46) D = yes
Christensen *et al.* (4)[42]	9 (♂)	Endurance-trained cyclists	64	3	BJ/150	9	Blackcurrant citrus with 0·2 mmol nitrate	1	180–249	Incremental arm exercise	Peak power output (W)	S = 140 (SD 17) NS = 141 (SD 20) D = no
Glaister *et al.*[60]	14 (♀)	Cyclists and triathletes	52·3	4·9	BJ/70	7·3	BJ negligible nitrate content	1	150	Time trial 20 km	Time (min)	S = 35·3 (SD 1·5) NS = 35·3 (SD 1·7) D = no
Hoon *et al.* (1)[61]	28 (♂)	Cyclists	–		BJ/70	4·1	BJ negligible nitrate content	1	75	Time trial 4 min	Mean power output (W)	S = 403 (SD 52) NS = 396 (SD 57) D = no

Table 2. *Continued*

			Nitrate supplementation									
			VO_{2peak}/ VO_{2max} (ml/kg per min)									
References	No. of subjects (♂, ♀)	Characteristics of subjects	Mean	SD	Ingested fluid/ volume (ml)	Dose (mmol)	Placebo substance	Days of supplementation	Time before trial (min)	Exercise protocol	Measure of physical performance	Results
Hoon *et al.* (2)[61]	28 (♂)	Cyclists	–		BJ/70	4·1	BJ negligible nitrate content	1	150	Time trial 4 min	Mean power output (W)	S = 402 (SD 47 NS = 396 (SD 57) D = no
Hoon *et al.* (1)[62]	10 (♂)	Rowers	–		BJ/70	4·2	BJ negligible nitrate content	1	120	Time trial 2000 m	Time (s)	S = 383·4 (SD 8·7) NS = 383·5 (SD 9) D = no
Hoon *et al.* (2)[62]	10 (♂)	Rowers	–		BJ/140	8·4	BJ negligible nitrate content	1	120	Time trial 2000 m	Time (s)	S = 381·9 (SD 9) NS = 383·5 (SD 9) D = yes
Kramer *et al.* (1)[63]	12 (♂)	CrossFit	48·5	7·0	Potassium nitrate	8·0	Nitrate-free potassium chloride	6	1440	Wingate test	Wingate peak	S = 948·0 (SD 186·8) NS = 905·0 (SD 157·2) D = yes
Kramer *et al.* (2)[63]	12 (♂)	CrossFit	48·5	7·0	Potassium nitrate	8·0	Nitrate-free potassium chloride	6	1440	Time trial 2 km	Time (s)	S = 459·7 (SD 23·9) NS = 459·8 (SD 24·8) D = no
Lane *et al.* (1)[64]	12 (♂)	Cyclists and triathletes	71·6	4·6	BJ/70	8·4	BJ negligible nitrate content	2	130	Time trial 43·83 km	Time (min)	S = 64·0 (SD 2·8) NS = 63·5 (SD 3·2) D = no
Lane *et al.* (2)[64]	12 (♂)	Cyclists and triathletes	71·6	4·6	BJ/70	8·4	BJ negligible nitrate content	2	130	Time trial 43·83 km	Power output (W)	S = 298 (SD 35 NS = 303 (SD 41) D = no
Lane *et al.* (3)[64]	12 (♀)	Cyclists and triathletes	59·9	5·1	BJ/70	8·4	BJ negligible nitrate content	2	130	Time trial 29·35 km	Time (min)	S = 51·6 (SD 2·6) NS = 51·6 (SD 2·5) D = no
Lane *et al.* (4)[64]	12 (♀)	Cyclists and triathletes	59·9	5·1	BJ/70	8·4	BJ negligible nitrate content	2	130	Time trial 29·35 km	Power output (W)	S = 207 (SD 31 NS = 207 (SD 29) D = no
Lansley *et al.* (1)[22]	9 (♂)	Cyclists	56·0	5·7	BJ/500	6·2	BJ negligible nitrate content	1	120	Time trial 4 km	Time (min)	S = 6·27 (SD 0·35) NS = 6·45 (SD 0·42) D = yes
Lansley *et al.* (2)[22]	9 (♂)	Cyclists	56·0	5·7	BJ/500	6·2	BJ negligible nitrate content	1	120	Time trial 4 km	Mean power output (W)	S = 292 (SD 44) NS = 279 (SD 51) D = yes
Lansley *et al.* (3)[22]	9 (♂)	Cyclists	56·0	5·7	BJ/500	6·2	BJ negligible nitrate content	1	120	Time trial 16·1 km	Time (min)	S = 26·9 (SD 1·8) NS = 27·7 (SD 2·1) D = yes
Lansley *et al.* (4)[22]	9 (♂)	Cyclists	56·0	5·7	BJ/500	6·2	BJ negligible nitrate content	1	120	Time trial 16·1 km	Mean power output (W)	S = 247 (SD 44 NS = 233 (SD 43) D = yes
Lowings *et al.*[65]	10 (5 ♂ and 5 ♀)	Swimmers	–		BJ/140 (70 + 70)	12·5	BJ negligible nitrate content	1	180	Swim time trial 168 m	Time (s)	S = 130·3 (SD 8·1) NS = 131·5 (SD 9·0) D = yes
Martin *et al.* (1)[66]	16 (9 ♂ and 7 ♀)	Team-sport players	47·2	8·5	BJ/70	4·83	BJ negligible nitrate content	1	120	8-s bouts of high-intensity intermittent-sprint test	No. of sprints completed	S = 13 (SD 5) NS = 15 (SD 6) D = yes
Martin *et al.* (2)[66]	16 (9 ♂ and 7 ♀)	Team-sport players	47·2	8·5	BJ/70	4·83	BJ negligible nitrate content	1	120	8-s bouts of high-intensity intermittent-sprint test	Work (kJ)	S = 49·2 (SD 24·2) NS = 57·8 (SD 34·0) D = yes
Martin *et al.* (3)[66]	16 (9 ♂ and 7 ♀)	Team-sport players	47·2	8·5	BJ/70	4·83	BJ negligible nitrate content	1	120	8-second bouts of high-intensity intermittent-sprint test	Mean power output (W)	S = 447 (SD 104) NS = 444 (SD 117) D = no
McQuillan *et al.* (1)[67]	9 (♂)	Cyclists	68	3	BJ/140	8·0	BJ negligible nitrate content	4	150	Time trial 1 km	Time (s)	S = 79·6 (SD 3·5) NS = 79·2 (SD 2·9) D = no
McQuillan *et al.* (2)[67]	9 (♂)	Cyclists	68	3	BJ/140	8·0	BJ negligible nitrate content	4	150	Time trial 1 km	Mean power output (W)	S = 495 (SD 61 NS = 503 (SD 51) D = no

Table 2. *Continued*

References	No. of subjects (♂, ♀)	Characteristics of subjects	Nitrate supplementation: VO_{2peak}/VO_{2max} (ml/kg per min) Mean	SD	Ingested fluid/ volume (ml)	Dose (mmol)	Placebo substance	Days of supplementation	Time before trial (min)	Exercise protocol	Measure of physical performance	Results
McQuillan *et al.* (3)[67]	9 (♂)	Cyclists	68	3	BJ/140	8·0	BJ negligible nitrate content	7	150	Time trial 1 km	Time (s)	S = 79·3 (SD 3·3) NS = 79·0 (SD 3·0) D = no
McQuillan *et al.* (4)[67]	9 (♂)	Cyclists	68	3	BJ/140	8·0	BJ negligible nitrate content	7	150	Time trial 1 km	Mean power output (W)	S = 501 (SD 59) NS = 505 (SD 52) D = no
McQuillan *et al.* (5)[67]	9 (♂)	Cyclists	68	3	BJ/140	8·0	BJ negligible nitrate content	3	150	Time trial 4 km	Time (s)	S = 341 (SD 12) NS = 340 (SD 10) D = no
McQuillan *et al.* (6)[67]	9 (♂)	Cyclists	68	3	BJ/140	8·0	BJ negligible nitrate content	3	150	Time trial 4 km	Mean power output (W)	S = 390 (SD 45) NS = 393 (SD 37) D = no
McQuillan *et al.* (7)[67]	9 (♂)	Cyclists	68	3	BJ/140	8·0	BJ negligible nitrate content	6	150	Time trial 4 km	Time (s)	S = 340 (SD 10) NS = 340 (SD 11) D = no
McQuillan *et al.* (8)[67]	9 (♂)	Cyclists	68	3	BJ/140	8·0	BJ negligible nitrate content	6	150	Time trial 4 km	Mean power output (W)	S = 394 (SD 38) NS = 393 (SD 37) D = no
McQuillan *et al.* (1)[68]	8 (♂)	Cyclists	63	4	BJ/70	4·0	BJ negligible nitrate content	8	120	Time trial 4 km	Time (s)	S = 343·6 (SD 14·3) NS = 344·8 (SD 14·0) D = no
McQuillan *et al.* (2)[68]	8 (♂)	Cyclists	63	4	BJ/70	4·0	BJ negligible nitrate content	8	120	Time trial 4 km	Mean power output	S = 380 (SD 41) NS = 375 (SD 40) D = no
Muggeridge *et al.* (1)[69]	8 (♂)	Kayakers	49·0	6·1	BJ/70	5·0	Tomato juice	1	180	Steady-state paddling at 60 % of WR_{max} (15 min)	Mean power output (W)	S = 108 (SD 64·8) NS = 108 (SD 62·0) D = no
Muggeridge *et al.* (2)[69]	8 (♂)	Kayakers	49·0	6·1	BJ/70	5·0	Tomato juice	1	180	Time trial 1 km	Time (s)	S = 276 (SD 14·1) NS = 277 (SD 14·1) D = no
Nyakayiru *et al.*[70]	17 (♂)	Cyclists and triathletes	65·0	4·0	Sodium nitrate/140	12·9	Sodium chloride	6	240	Time trial 10 km	Time (s)	S = 1004 (SD 67) NS = 1017 (SD 71) D = no
Peacock *et al.*[28]	10 (♂)	Elite cross-country skiers	69·6	5·1	1 g of potassium nitrate in a capsule	9·9	1 g of maltodextrin in a capsule	1	150	Time trial 5 km	Time (s)	S = 1005 (SD 53) NS = 996 (SD 49) D = no
Peeling *et al.* (1)[71]	6 (♂)	Kayakers	57·15	2·77	BJ/70	4·8	BJ negligible nitrate content	1	150	4-min all-out maximal effort on the stationary kayak ergometer	Power output (W)	S = 319 (SD 35) NS = 318 (SD 42) D = no
Peeling *et al.* (2)[71]	6 (♂)	Kayakers	57·15	2·77	BJ/70	4·8	BJ negligible nitrate content	1	150	4-min all-out maximal effort on the stationary kayak ergometer	Distance covered (m)	S = 989 (SD 31) NS = 982 (SD 36) D = no
Peeling *et al.* (3)[71]	5 (♀)	Kayakers	47·8	3·7	BJ/70	9·6	BJ negligible nitrate content	1	120	Time trial 500 m	Time (s)	S = 114·6 (SD 1·5) NS = 116·7 (SD 2·2) D = yes
Peeling *et al.* (4)[71]	5 (♀)	Kayakers	47·8	3·7	BJ/70	9·6	BJ negligible nitrate content	1	120	Time trial 500 m	Velocity in 100–400 m (m/s)	S = 4·4 (SD 0·03) NS = 4·3 (SD 0·05) D = yes
Rimer *et al.* (1)[72]	13 (11 ♂ and 2 ♀)	Tennis, Alpine Ski, American Football, Cycling, Triathlon	–		BJ/140 (70 + 70)	11·2	BJ negligible nitrate content	1	150	4×, maximal inertial-load cycling trial (3-4 s)	Maximal power output (W)	S = 1229 (SD 317) NS = 1213 (SD 300) D = yes
Rimer *et al.* (2)[72]	13 (11 ♂ and 2 ♀)	Tennis, Alpine Ski, American Football, Cycling, Triathlon	–		BJ/140 (70 + 70)	11·2	BJ negligible nitrate content	1	150	Maximal isokinetic cycling trial, 120 rpm (30 s)	Total work (kJ)	S = 22·8 (SD 4·8) NS = 23·0 (4·4) D = no
Rimer *et al.*[73]	13 (11 ♂ and 2 ♀)	Tennis, Alpine Ski, American Football, Cycling, Triathlon	–		BJ/140 (70 + 70)	11·2	BJ negligible nitrate content	1	150	Maximal isokinetic cycling trial, 120 rpm (30 s)	Peak Power (W)	S = 1173 (SD 255) NS = 1185 (SD 249) D = no

Table 2. *Continued*

References	No. of subjects (♂, ♀)	Characteristics of subjects	Nitrate supplementation: VO2peak/VO2max (ml/kg per min) Mean	SD	Ingested fluid/volume (ml)	Dose (mmol)	Placebo substance	Days of supplementation	Time before trial (min)	Exercise protocol	Measure of physical performance	Results
Shannon *et al.* (1)[74]	8 (♂)	Runners or triathletes	62·3	8·1	BJ/140	12·5	BJ negligible nitrate content	1	180	Time trial 1·500 m	Time (s)	S = 319·6 (SD 36·2) NS = 325·7 (SD 38·8) D = yes
Shannon *et al.* (2)[74]	8 (♂)	Runners or triathletes	62·3	8·1	BJ/140	12·5	BJ negligible nitrate content	1	180	Time trial 10·000 m	Time (s)	S = 2643·1 (SD 324·1) NS = 2649·9 (SD 319·8) D = no
Thompson *et al.* (1)[75]	36 (♂)	Team-sport players	–		BJ/70	6·4	BJ negligible nitrate content	5	150	Sprints (5 × 20 m)	Time (s) at 20 m	S = 3·98 (SD 0·18) NS = 4·03 (SD 0·19) D = yes
Thompson *et al.* (2)[75]	36 (♂)	Team-sport players	–		BJ/70	6·4	BJ negligible nitrate content	5	150	Teste Yo-Yo IR1 (2 × 20 m)	Distance covered (m)	S = 1422 (SD 502) NS = 1369 (SD 505) D = yes
Wilkerson *et al.* (1)[76]	8 (♂)	Cyclists	63	8	BJ/500	6·2	BJ negligible nitrate content	1	150	Time trial 50 miles	Time (min)	S = 136·7 (SD 5·6) NS = 137·9 (SD 6·4) D = no
Wilkerson *et al.* (2)[76]	8 (♂)	Cyclists	63	8	BJ/500	6·2	BJ negligible nitrate content	1	150	Time trial 50 miles	Mean power output (W)	S = 238 (SD 22) NS = 235 (SD 27) D = no

♂, Male; ♀, female; BJ, beetroot juice; NR, not reported; S, supplemented; NS, no supplementation; D, statistical difference.

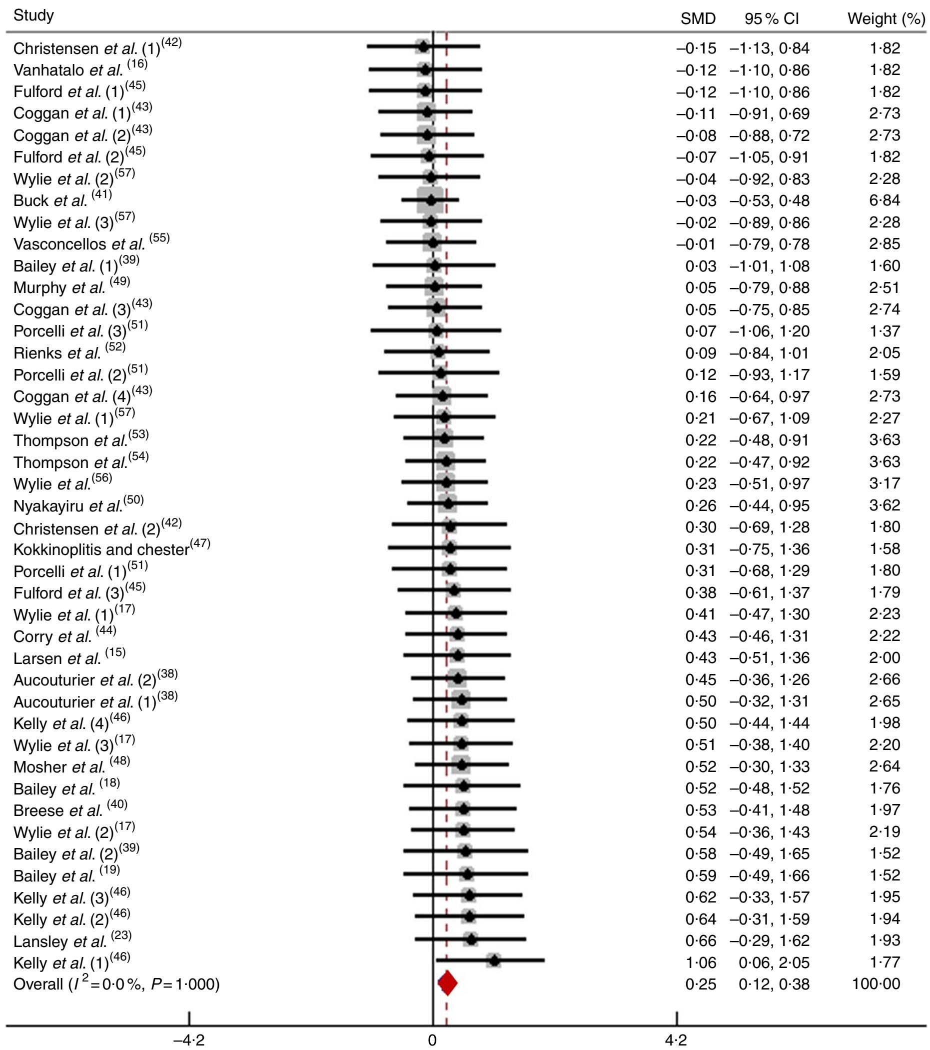

Fig. 3. Forest plot of physical performance following dietary NO_3^- supplementation in non-athletes. SMD, standardised mean difference.

analysis, heterogeneity was observed among these studies ($I^2 = 0\,\%$; $Q = 13{\cdot}31$, df = 16, $P = 0{\cdot}65$).

Non-athletes subjected to long-duration tests. After pooling the data from twenty-five trials, the mean effect size was 0·33 (95 % CI 0·15, 0·51), which indicates that the dietary NO_3^- supplementation had a small and significant beneficial effect on physical performance ($P < 0{\cdot}05$; Fig. 7). According to a fixed-effects analysis, no heterogeneity was observed among these studies ($I^2 = 0\,\%$; $Q = 8{\cdot}01$, df = 24, $P = 0{\cdot}99$).

Athletes subjected to long-duration tests. After pooling the data from forty-four trials, the mean effect size was 0·05 (95 % CI –0·07, 0·17), which indicates that the dietary NO_3^- supplementation had a negligible and non-significant effect on physical performance ($P > 0{\cdot}05$; Fig. 8). According to a fixed-effects analysis, no heterogeneity was observed among these studies ($I^2 = 0\,\%$; $Q = 4{\cdot}82$, df = 43, $P = 1{\cdot}00$). The subsequent analysis consisted of subdividing the non-athletes that performed long-duration tests according to the test protocol used.

Non-athletes subjected to long-duration, open-ended tests. After pooling the data from fourteen trials, the mean effect size was 0·47 (95 % CI 0·23, 0·71), which indicates that the dietary NO_3^- supplementation had a small and significant beneficial effect on physical performance ($P < 0{\cdot}05$; Fig. 9). According to a fixed-effects analysis, no heterogeneity was observed among these studies ($I^2 = 0\,\%$; $Q = 3{\cdot}77$, df = 13, $P = 0{\cdot}99$).

Non-athletes subjected to long-duration time trials. After pooling the data from four trials, the mean effect size was 0·12 (95 % CI –0·37, 0·61), which indicates that the dietary NO_3^- supplementation had a negligible and non-significant effect on physical performance ($P > 0{\cdot}05$; Fig. 10). According to a fixed-effects analysis, no heterogeneity was observed among these studies ($I^2 = 0\,\%$; $Q = 0{\cdot}16$, df = 3, $P = 0{\cdot}98$).

Non-athletes subjected to long-duration, graded-exercise tests. After pooling the data from five trials, the mean effect size was 0·20 (95 % CI –0·18, 0·59), which indicates that the dietary NO_3^- supplementation had a small but non-significant effect on

British Journal of Nutrition

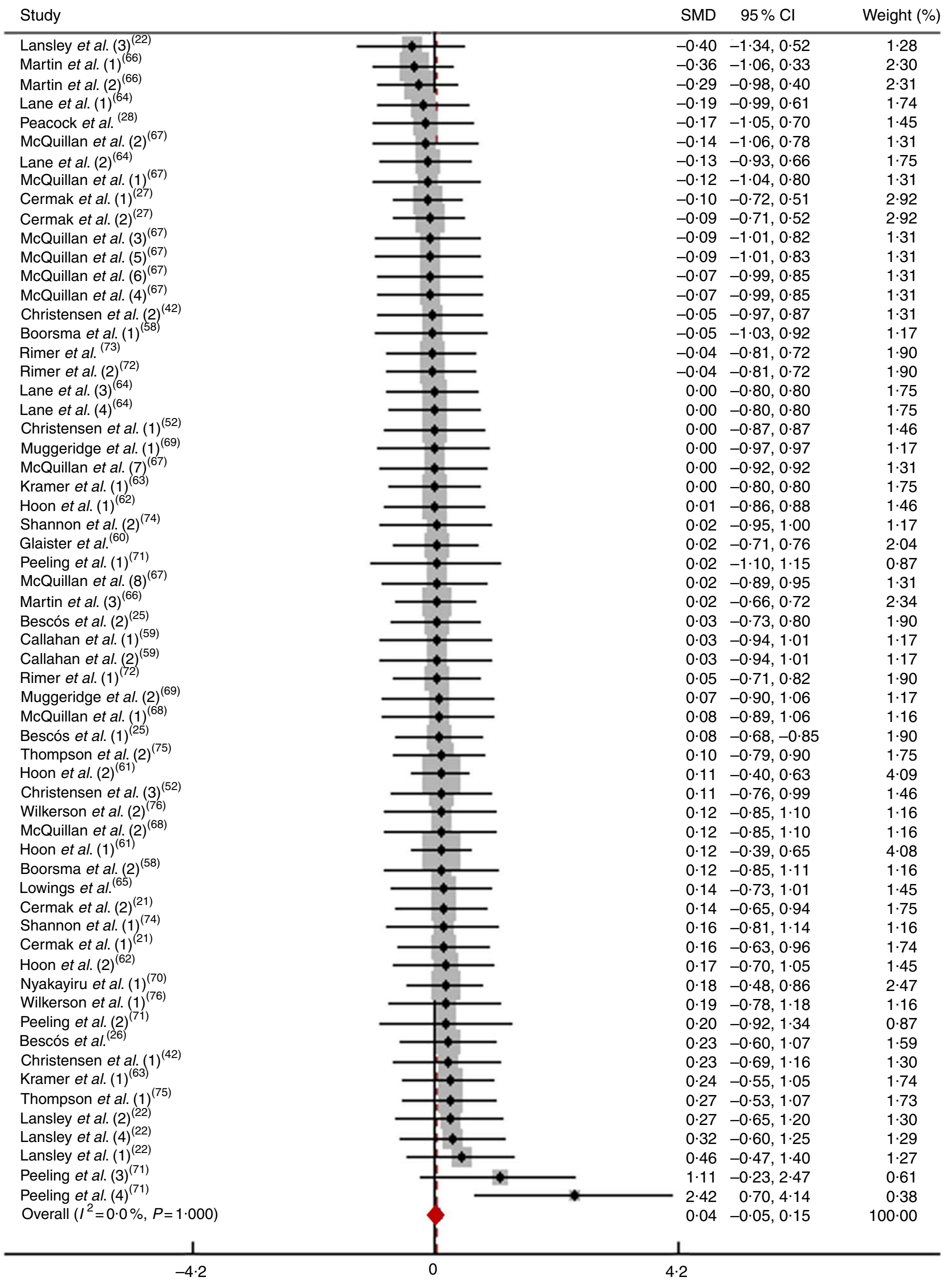

Fig. 4. Forest plot of physical performance following dietary NO_3^- supplementation in athletes. SMD, standardised mean difference.

physical performance ($P > 0{\cdot}05$; Fig. 11). According to a fixed-effects analysis, no heterogeneity was observed among these studies ($I^2 = 0\,\%$; $Q = 1{\cdot}38$, df $= 4$, $P = 0{\cdot}84$).

Cyclists. Most tested athletes were cyclists; therefore, this subgroup was subjected to a special analysis in which they were evaluated alone without the inclusion of athletes engaged in other sports. After pooling the data from thirty-seven trials, the effect size mean was 0·04 (95 % CI –0·09, 0·17), which indicates that the dietary NO_3^- supplementation had a negligible and non-significant effect on physical performance ($P > 0{\cdot}05$; Fig. 12). According to a fixed-effects analysis, heterogeneity was observed among these studies ($I^2 = 0\,\%$; $Q = 4{\cdot}90$, df $= 36$, $P = 1{\cdot}00$).

Analysis of the relationship between the performance level and the ergogenic response to the NO_3^- supplementation

By analysing the percentage of trials reporting increased performance in individuals classified into different PL, we observed numerous trials, that is, 50 and 56·5 %, showing increased performance in individuals with PL1 and PL2, respectively. In contrast, approximately 37 % of the trials involving individuals with PL3 showed an increased performance following the NO_3^- supplementation, whereas in trials involving individuals with PL4 and PL5 no improvement in performance was observed following the NO_3^- supplementation (Fig. 13). The χ^2 test showed a different distribution among the PL ($P = 0{\cdot}002$).

British Journal of Nutrition

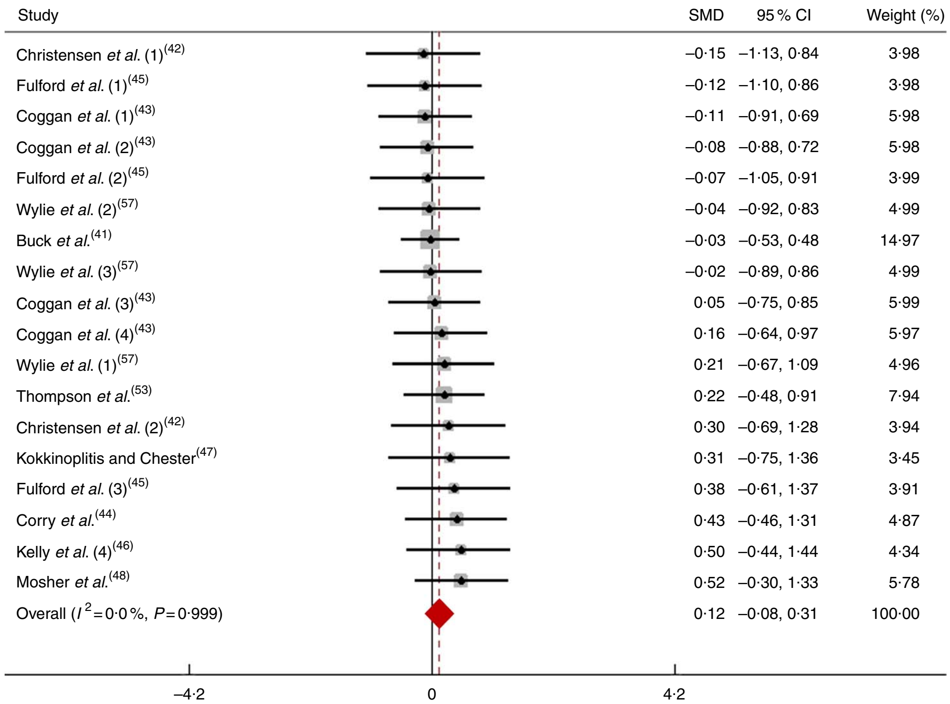

Fig. 5. Forest plot of physical performance during a short-duration test following dietary NO_3^- supplementation in non-athletes. SMD, standardised mean difference.

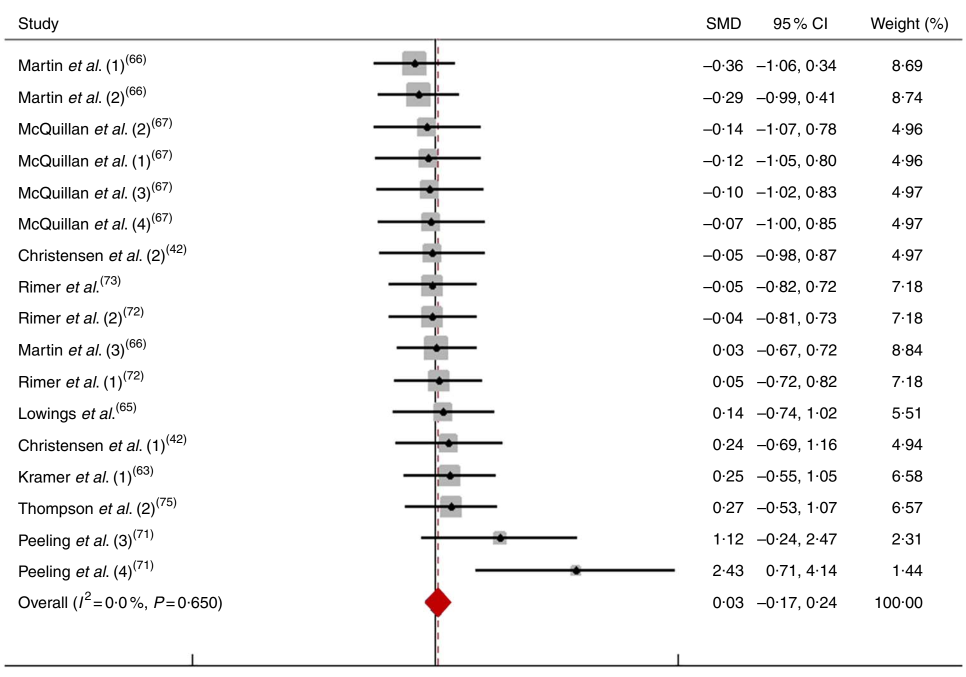

Fig. 6. Forest plot of physical performance during a short-duration test following dietary NO_3^- supplementation in athletes. SMD, standardised mean difference.

Association between supplementation features and changes in physical performance

Pearson's correlation analyses were performed to verify the association between these variables, including the association between changes in physical performance and the dose of NO_3^- (non-athletes: r 0·351, $P>0·05$; athletes: r 0·099, $P>0·05$), the number of days of supplementation (non-athletes: r 0·166, $P>0·05$; athletes: r 0·114, $P>0·05$) and the total amount ingested (dose multiplied by days under supplementation) (non-athletes: r 0·112, $P>0·05$; athletes: r 0·088, $P>0·05$). No significant correlations were observed between the supplementation features evaluated and changes in physical performance.

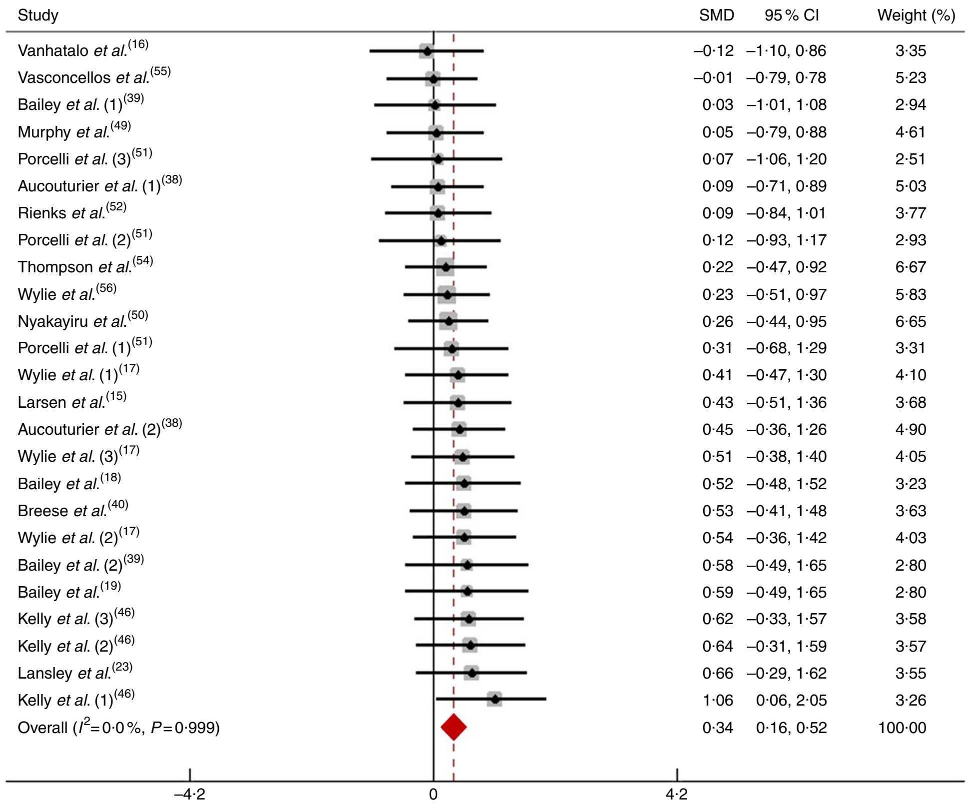

Fig. 7. Forest plot of physical performance during a long-duration test following dietary NO_3^- supplementation in non-athletes. SMD, standardised mean difference.

Publication bias

Publication bias was assessed by a visual inspection of the funnel plot for all subgroups analysed: non-athletes (online Supplementary Fig. S2(a)), athletes (online Supplementary Fig. S1(a)), non-athletes subjected to short-duration tests (online Supplementary Fig. S2(b)), athletes subjected to short-duration tests (online Supplementary Fig. S1(b)), non-athletes subjected to long-duration tests (online Supplementary Fig. S2(c)), athletes subjected to long-duration tests (online Supplementary Fig. S1 (c)), non-athletes subjected to long-duration, open-ended tests (online Supplementary Fig. S3(a)), non-athletes subjected to long-duration time trials (online Supplementary Fig. S3(b)) and non-athletes subjected to long-duration, graded-exercise tests (online Supplementary Fig. S3(c)). These analyses revealed minor asymmetrical inverted distributions that were prominent in all plots, suggesting the presence of a small publication bias.

Risk of bias

The risk of bias was assessed in fifty-four studies (twenty-six and twenty-eight conducted with non-athletes and athletes, respectively) in the systematic review. One study[71] was subjected to two independent evaluations because it presented independent experimental trials. Out of fifty-five evaluations, forty-eight did not present any major risk of bias. Approximately 13 % (non-athletes, two studies; athletes, five studies) of the studies did not blind the participants or researchers. In general, the studies evaluated in the present systematic review showed consistent control of the risk of bias and were deemed to be good-quality studies (online Supplementary Tables S3 and S4).

Discussion

The present systematic review and meta-analysis demonstrated that the level of physical fitness is a determining factor in the performance-enhancing effects associated with NO_3^- supplementation. Although athletes are usually less prone to benefit from NO_3^- supplementation, non-athletes can experience small but significant advantages in their physical performance, particularly in performance evaluations using long-duration, open-ended tests. Interestingly, this effect is not observed using time trials, which is the most ecologically valid exercise protocol[77]. These findings regarding the beneficial effects induced by NO_3^- supplementation in non-athletes are supported by the analysis in which the participants were subdivided according to their PL, and those classified at the lower levels (less conditioned) showed more improvements. This information is very important for exercise practitioners and athletes and provides support in decisions regarding whether to use this potential ergogenic aid to improve physical performance and health.

In the present meta-analysis, we observed that individuals with higher fitness levels benefit less from NO_3^- supplementation (Fig. 13). Consistently, the effect size of NO_3^- supplementation-mediated changes on performance in athletes was mostly irrelevant (Fig. 4). In contrast, non-athletes can benefit from NO_3^- supplementation (Fig. 3). This was the first study to systematically show the importance of characterising the fitness levels of

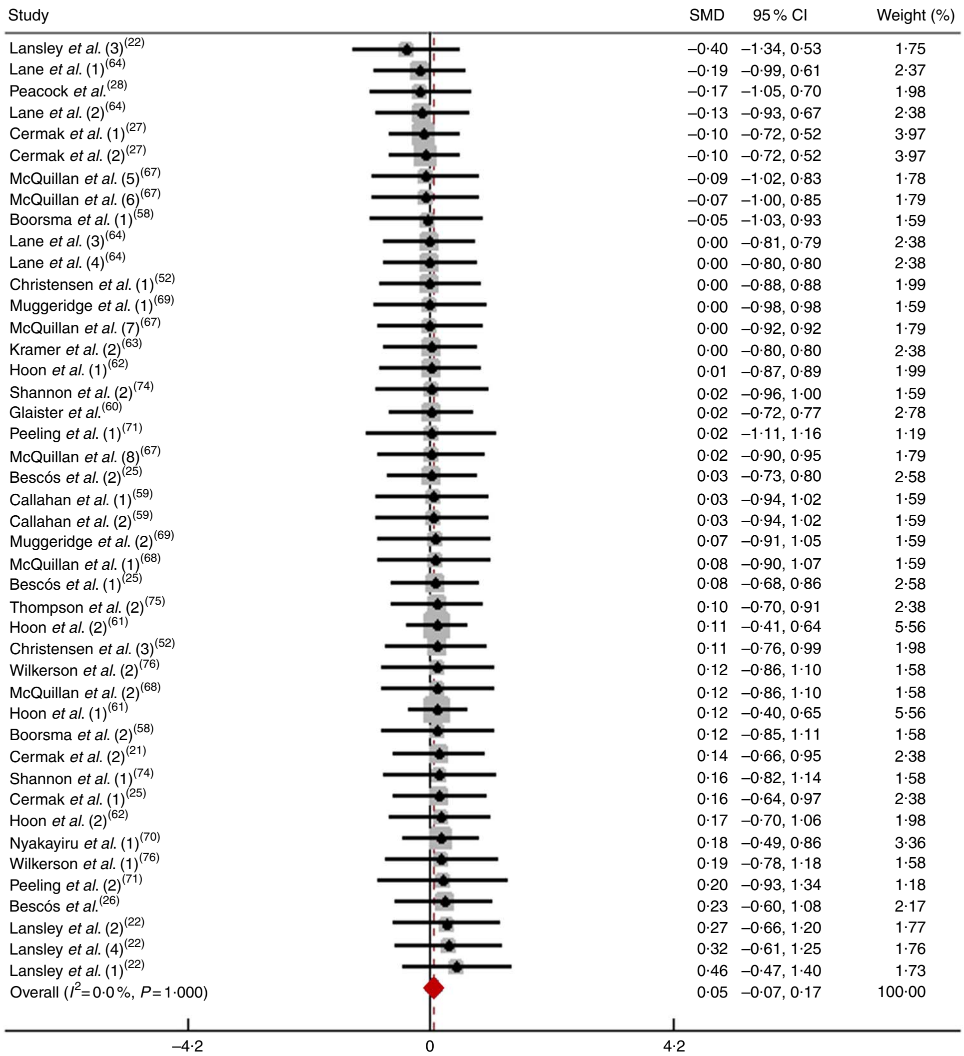

Fig. 8. Forest plot of physical performance during a long-duration test following dietary NO_3^- supplementation in athletes. SMD, standardised mean difference.

individuals before adopting a nutritional NO_3^- supplementation ergogenic strategy. Similarly, Porcelli *et al.*[51] assessed athletic performance in subjects with three aerobic fitness levels after 6 d of supplementation with 5·5 mmol per d of NO_3^-. The authors observed that individuals with lower and moderate aerobic capacities performed better during the time trial after the NO_3^- supplementation. However, the performance during the time trial was not improved in individuals with a higher aerobic capacity.

Several mechanisms may act collectively to improve performance following NO_3^- supplementation in non-athletes, including beneficial effects of an increased NO bioavailability in the skeletal muscles, blood vessels and even in the brain (Fig. 14). In contrast, the mechanisms underlying the limited ergogenic effects of NO_3^- supplementation in high-performance athletes have not been well elucidated. The ergogenic effects of NO_3^- supplementation are related to enhanced NO bioavailability, and athletes probably already have optimal levels of NO[51]. Highly trained subjects are likely to have high NOS activity[83], which might render the NO_3^-–NO_2^-–NO pathway less important for NO production. Therefore, the resulting increase in NO bioavailability due to supplementation does not appear to be relevant in athletes. In addition to these factors, Porcelli *et al.*[51] suggested that high-performance athletes have a high daily energy expenditure and possibly an enriched diet. Therefore, a diet consisting of a higher intake of NO_3^- in these subjects should be considered. Furthermore, recent evidence that NO_3^- supplementation may preferentially alter contractile function in type II fibres[79] suggests that endurance athletes, who typically have a low proportion of such fibres in their musculature[84], might experience a blunted physiological response to NO_3^- supplementation.

The effects of NO_3^- supplementation on exercise performance in non-athletes appear to be more robust in evaluations using long-duration, open-ended tests rather than time trials. Time-trial tests are the most ecologically valid options to assess performance[6,85]. Compared with time trials, constant-power (open-ended) tests are more influenced by psychological factors, such as boredom and motivation[86,87]. In addition, open-ended tests are more efficient in measuring endurance capacity rather than exercise performance, which is best measured by time-trial protocols[6,88].

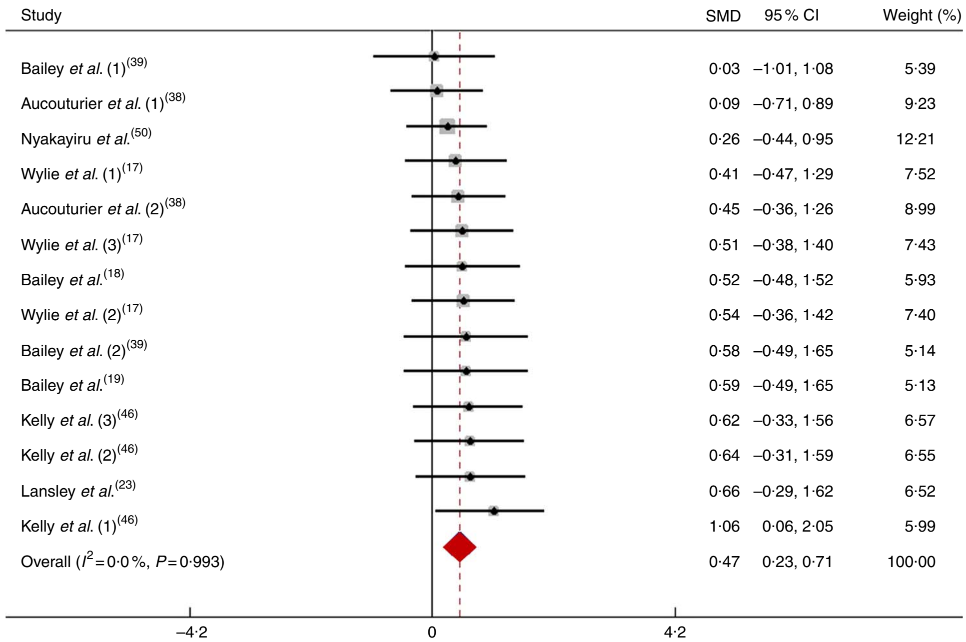

Fig. 9. Forest plot of physical performance during a long-duration open-ended test following dietary NO_3^- supplementation in non-athletes. SMD, standardised mean difference.

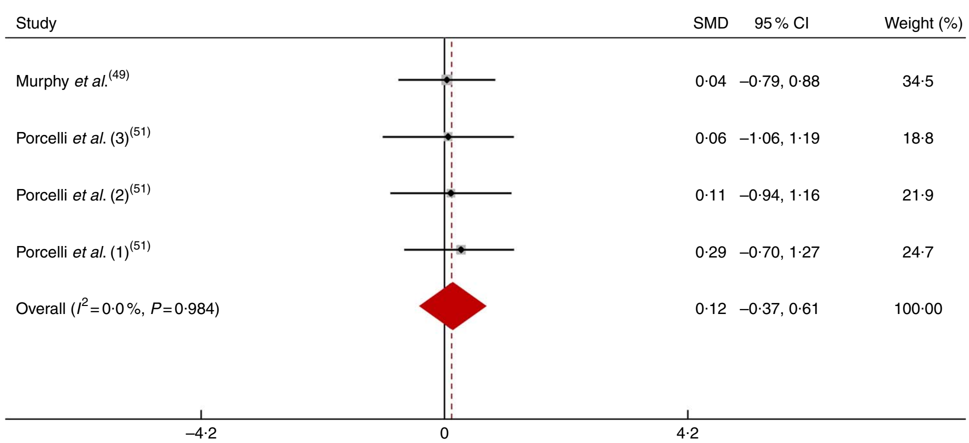

Fig. 10. Forest plot of physical performance during a long-duration time trial following dietary NO_3^- supplementation in non-athletes. SMD, standardised mean difference.

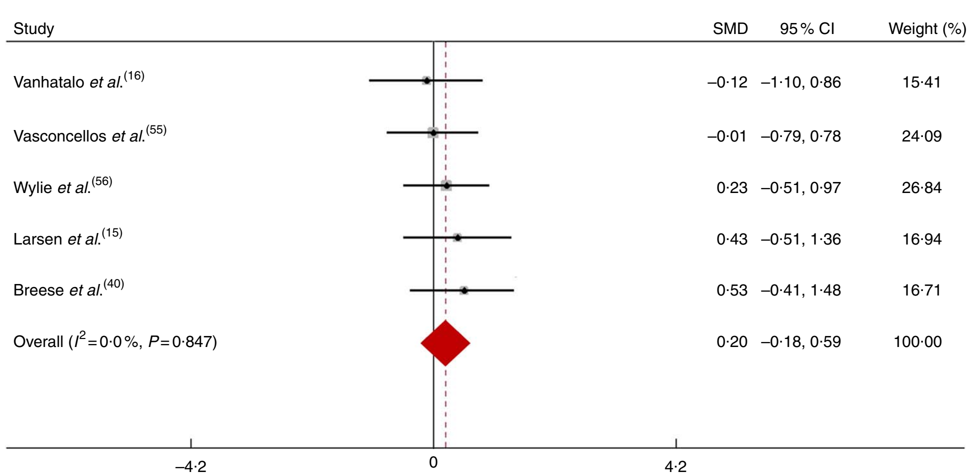

Fig. 11. Forest plot of physical performance during a long-duration graded-exercise test following dietary NO_3^- supplementation in non-athletes. SMD, standardised mean difference.

https://doi.org/10.1017/S0007114518000132 Published online by Cambridge University Press

British Journal of Nutrition

https://doi.org/10.1017/S0007114518000132 Published online by Cambridge University Press

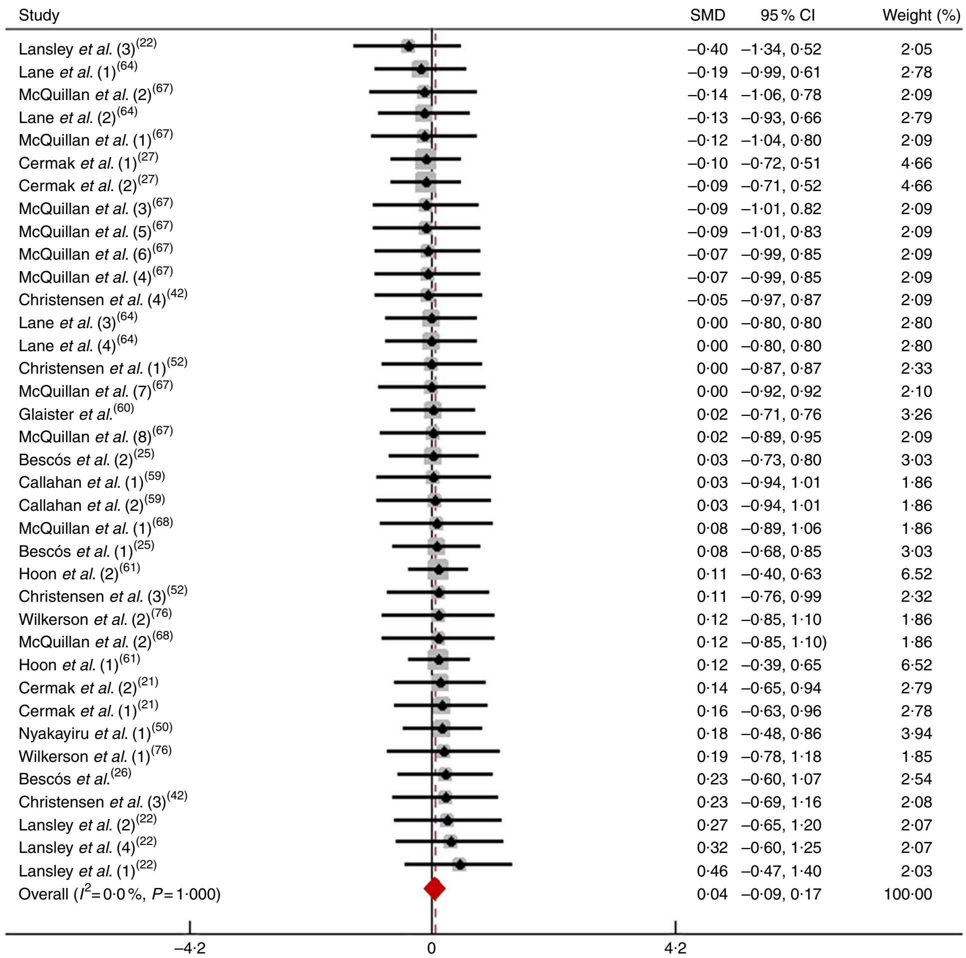

Fig. 12. Forest plot of physical performance in cyclists following dietary NO_3^- supplementation. SMD, standardised mean difference.

Although the dietary NO_3^- supplementation did not exert positive effects on the performance of athletes as previously described, the use of this supplement in sports competitions may still be applicable. During competitions, the winner is often determined by narrow differences between athletes, thus creating opportunities for the implementation of practices that may have subtle improvements in performance. Therefore, the distinct sensitivity of different athletes to supplementation should not be disregarded[7,76] and further research on this topic is warranted.

It is important to understand the physiological meaning of the doses that were supplemented in the included studies. These doses ranged from 4·0 to 19·5 mmol (Tables 1 and 2). Considering that the daily ingestion of NO_3^- corresponds on average to 91 mg (1·5 mmol) in people from the UK[89], the supplementation would increase the daily ingestion of nitrate by 3- to 13-fold in this population. However, the dose of the NO_3^- supplementation, the number of days of supplementation and the total amount ingested do not appear to influence the effects of NO_3^- supplementation on physical performance in non-athletes and athletes as shown by the lack of significant associations between these parameters. Studies using a single dose showed that NO_3^- supplementation had either no effects[41,43,45,47] or positive effects[17,53,56] on exercise performance. Likewise,

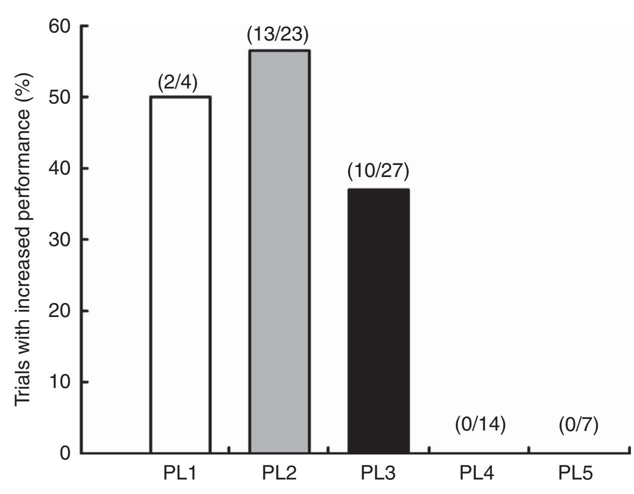

Fig. 13. Number of trials with increased performance (%) in subjects with different performance levels (PL).

studies using several days (≥5 d) of supplementation showed that NO_3^- supplementation had either no effects[19,39,45,57] or positive effects[18,22,40,46] on exercise performance. A similar rationale can be applied to the supplementation dose, which does not appear to influence physical performance.

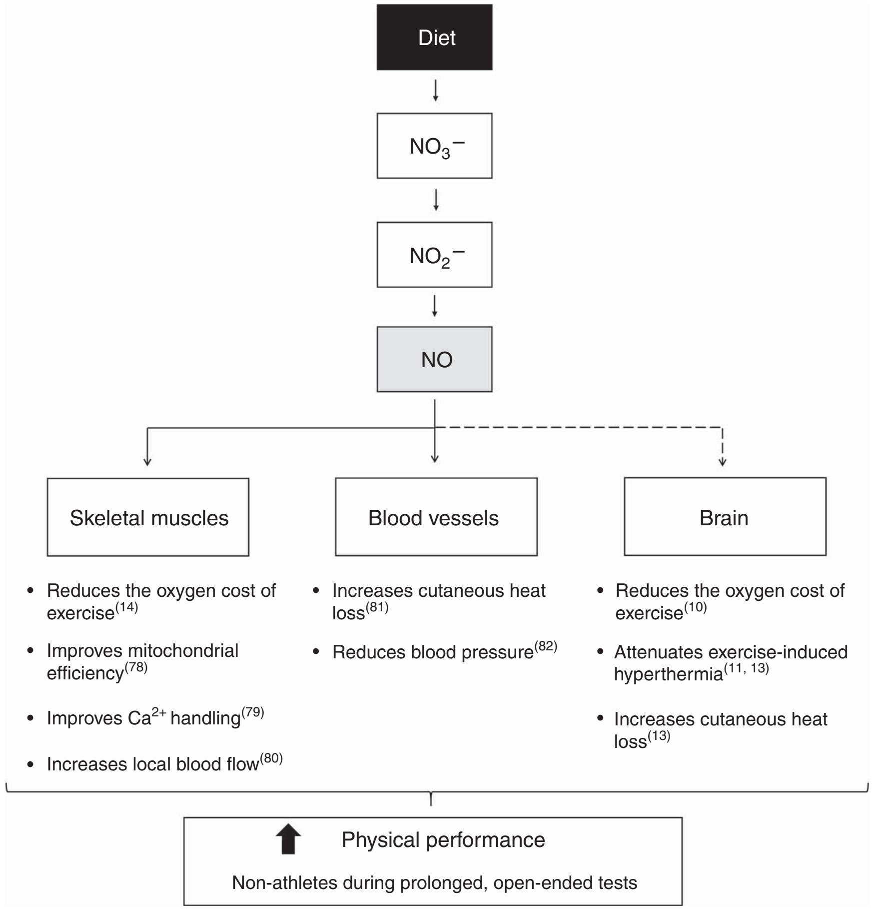

Fig. 14. Mechanisms underlying improved physical performance induced by nitrate (NO_3^-) supplementation in non-athletes subjected to prolonged, open-ended tests. Through a series of reduction reactions along the gastrointestinal tract and at target tissues, NO_3^- acts as the main nitric oxide (NO) donor. Increased NO bioavailability promotes beneficial effects on performance through effects in skeletal muscles, blood vessels and likely in the brain. To date, no study has provided direct evidence showing that NO_3^- supplementation increases brain NO levels (this is the reason why a dashed line is connecting NO to the brain in the schematic). In the skeletal muscles, NO reduces the oxygen cost of exercise[14], improves mitochondrial efficiency[78] and Ca^{2+} handling[79] and increases local blood flow[80]. In the blood vessels, NO increases cutaneous heat loss[81] and reduces blood pressure[82]. Experiments conducted in rats showed that NO in the brain reduces the oxygen cost of exercise[10], attenuates exercise-induced hyperthermia[11,13] and increases cutaneous heat loss[13]. Collectively, these physiological responses induced by NO_3^- supplementation improve performance in the conditions mentioned above.

For example, studies using low doses (4–5·5 mmol) showed that NO_3^- supplementation had either no effects[17,19,38,51] or positive effects[16,51,53,56] on exercise performance. Finally, studies using high doses (>10 mmol) also showed that NO_3^- supplementation had no effects[43,45,52] or positive effects[17,43] on exercise performance.

Despite the high variability in the experimental protocols used in the studies analysed in the present review, the analysed subgroups did not include heterogeneous samples. Therefore, the data homogeneity, the quality of the studies assessed by the risk of bias and the absence of publication bias in the studies used in this systematic review and meta-analysis are sufficient to draw conclusions.

A major limitation of this review is related to the wide variation in the methods (differences in the dose of NO_3^-, number of days of supplementation, total amount ingested and mode of NO_3^- delivery) used in the analysed studies. This methodological diversity complicates the interpretation of the results and precludes clear conclusions regarding certain features of supplementation, such as those listed above. In addition, most studied individuals were men, and whether a sex-related sensitivity to the enhancing effects of nitrate exists in non-athletes is unclear. Thus, future studies should include women as participants.

Practical applications

The present results may encourage coaches, athletes and exercise practitioners to consider the following: (1) NO_3^- supplementation appears to be more effective in non-athletes than in athletes, particularly in performance evaluations using long-duration, open-ended tests; (2) the ergogenic effects mediated by NO_3^- supplementation do not affect physical performance in athletes, including cyclists, which are the most studied athletic population; and (3) subjects classified at a lower PL (i.e. less conditioned) are more responsive to the effects of NO_3^- supplementation than are subjects classified at a higher PL.

Conclusion

The present systematic review and meta-analysis indicates that dietary NO_3^- supplementation improves physical performance in non-athletes, particularly in performance evaluations using long-duration, open-ended tests. In contrast, dietary NO_3^- supplementation does not appear to benefit the performance of athletes.

Acknowledgements

The authors thank Dr Luc Van Loon for his helpful comments to the first draft of this manuscript.

This study was supported by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) and Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG). The funding institutions had no role in the study design, data analysis, decision to publish or preparation of the article.

All authors contributed to the development of the research question and study design. H. O. C., L. R. D., Q. T. R. and W. P. performed the literature search. H. O. C., L. R. D., Q. T. R. and F. S. M. M. performed the study selection. H. O. C., L. R. D. and Q. T. R. analysed the data. All authors interpreted the results and wrote the manuscript. All authors reviewed and approved the manuscript.

The authors declare that there are no conflicts of interest.

Supplementary material

For supplementary material/s referred to in this article, please visit http://dx.doi.org/10.1017/S0007114518000132

References

1. Jones AM, Haramizu S, Ranchordas M, *et al.* (2011) A–Z of nutritional supplements: dietary supplements, sports nutrition foods and ergogenic aids for health and performance – part 27. *Br J Sports Med* **45**, 1246–1248.
2. Jones AM, Vanhatalo A & Bailey SJ (2013) Influence of dietary nitrate supplementation on exercise tolerance and performance. *Nestle Nutr Inst Workshop Ser* **75**, 27–40.
3. Jones AM (2014) Dietary nitrate supplementation and exercise performance. *Sports Med* **44**, Suppl. 1, S35–S45.
4. Thompson C, Wylie LJ, Blackwell JR, *et al.* (2017) Influence of dietary nitrate supplementation on physiological and muscle metabolic adaptations to sprint interval training. *J Appl Physiol (1985)* **122**, 642–652.
5. Hoon MW, Johnson NA, Chapman PG, *et al.* (2013) The effect of nitrate supplementation on exercise performance in healthy individuals: a systematic review and meta-analysis. *Int J Sport Nutr Exerc Metab* **23**, 522–532.
6. McMahon NF, Leveritt MD & Pavey TG (2017) The effect of dietary nitrate supplementation on endurance exercise performance in healthy adults: a systematic review and meta-analysis. *Sports Med* **47**, 735–756.
7. Jonvik KL, Nyakayiru J, van Loon LJ, *et al.* (2015) Last word on viewpoint: can elite athletes benefit from dietary nitrate supplementation? *J Appl Physiol (1985)* **119**, 770.
8. Jonvik KL, Nyakayiru J, van Loon LJ, *et al.* (2015) Can elite athletes benefit from dietary nitrate supplementation? *J Appl Physiol (1985)* **119**, 759–761.
9. Hultstrom M, Amorim de Paula C, Antonio Peliky Fontes M, *et al.* (2015) Commentaries on viewpoint: can elite athletes benefit from dietary nitrate supplementation? *J Appl Physiol (1985)* **119**, 762–769.
10. Lacerda AC, Marubayashi U, Balthazar CH, *et al.* (2006) Evidence that brain nitric oxide inhibition increases metabolic cost of exercise, reducing running performance in rats. *Neurosci Lett* **393**, 260–263.
11. Lacerda AC, Marubayashi U & Coimbra CC (2005) Nitric oxide pathway is an important modulator of heat loss in rats during exercise. *Brain Res Bull* **67**, 110–116.
12. Lima PM, Santiago HP, Szawka RE, *et al.* (2014) Central blockade of nitric oxide transmission impairs exercise-induced neuronal activation in the PVN and reduces physical performance. *Brain Res Bull* **108**, 80–87.
13. Wanner SP, Leite LH, Guimaraes JB, *et al.* (2015) Increased brain L-arginine availability facilitates cutaneous heat loss induced by running exercise. *Clin Exp Pharmacol Physiol* **42**, 609–616.
14. Larsen FJ, Weitzberg E, Lundberg JO, *et al.* (2007) Effects of dietary nitrate on oxygen cost during exercise. *Acta Physiol* **191**, 59–66.
15. Larsen FJ, Weitzberg E, Lundberg JO, *et al.* (2010) Dietary nitrate reduces maximal oxygen consumption while maintaining work performance in maximal exercise. *Free Radic Biol Med* **48**, 342–347.
16. Vanhatalo A, Bailey SJ, Blackwell JR, *et al.* (2010) Acute and chronic effects of dietary nitrate supplementation on blood pressure and the physiological responses to moderate-intensity and incremental exercise. *Am J Physiol Regul Integr Comp Physiol* **299**, R1121–R1131.
17. Wylie LJ, Kelly J, Bailey SJ, *et al.* (2013) Beetroot juice and exercise: pharmacodynamic and dose–response relationships. *J Appl Physiol (1985)* **115**, 325–336.
18. Bailey SJ, Winyard P, Vanhatalo A, *et al.* (2009) Dietary nitrate supplementation reduces the O_2 cost of low-intensity exercise and enhances tolerance to high-intensity exercise in humans. *J Appl Physiol* **107**, 1144–1155.
19. Bailey SJ, Fulford J, Vanhatalo A, *et al.* (2010) Dietary nitrate supplementation enhances muscle contractile efficiency during knee-extensor exercise in humans. *J Appl Physiol* **109**, 135–148.
20. Bond H, Morton L & Braakhuis AJ (2012) Dietary nitrate supplementation improves rowing performance in well-trained rowers. *Int J Sport Nutr Exerc Metab* **22**, 251–256.
21. Cermak NM, Gibala MJ & van Loon LJC (2012) Nitrate supplementation's improvement of 10-km time-trial performance in trained cyclists. *Int J Sport Nutr Exerc Metab* **22**, 64–71.
22. Lansley KE, Winyard PG, Bailey SJ, *et al.* (2011) Acute dietary nitrate supplementation improves cycling time trial performance. *Med Sci Sports Exerc* **43**, 1125–1131.
23. Lansley KE, Winyard PG, Fulford J, *et al.* (2011) Dietary nitrate supplementation reduces the O_2 cost of walking and running: a placebo-controlled study. *J Appl Physiol* **110**, 591–600.
24. Masschelein E, Van Thienen R, Wang X, *et al.* (2012) Dietary nitrate improves muscle but not cerebral oxygenation status during exercise in hypoxia. *J Appl Physiol (1985)* **113**, 736–745.
25. Bescós R, Ferrer-Roca V, Galilea PA, *et al.* (2012) Sodium nitrate supplementation does not enhance performance of endurance athletes. *Med Sci Sports Exerc* **44**, 2400–2409.
26. Bescós R, Rodriguez FA, Iglesias X, *et al.* (2011) Acute administration of inorganic nitrate reduces VO(2peak) in endurance athletes. *Med Sci Sports Exerc* **43**, 1979–1986.

27. Cermak NM, Res P, Stinkens R, *et al.* (2012) No improvement in endurance performance after a single dose of beetroot juice. *Int J Sport Nutr Exerc Metab* **22**, 470–478.
28. Peacock O, Tjonna AE, James P, *et al.* (2012) Dietary nitrate does not enhance running performance in elite cross-country skiers. *Med Sci Sports Exerc* **44**, 2213–2219.
29. Wilkerson DP, Hayward G, Stephen BJ, *et al.* (2012) Acute dietary nitrate supplementation does not improve 50-mile time trial performance in highly trained cyclists. *Med Sci Sport Exerc* **44**, 442–442.
30. Liberati A, Altman DG, Tetzlaff J, *et al.* (2009) The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate health care interventions: explanation and elaboration. *Ann Intern Med* **151**, W65–W94.
31. Moher D, Liberati A, Tetzlaff J, *et al.* (2009) Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *Ann Intern Med* **151**, 264–269, W264.
32. Christensen PM, Nyberg M & Bangsbo J (2013) Influence of nitrate supplementation on VO_2 kinetics and endurance of elite cyclists. *Scand J Med Sci Sports* **23**, e21–e31.
33. Duffield R, Dawson B & Goodman C (2005) Energy system contribution to 400-metre and 800-metre track running. *J Sports Sci* **23**, 299–307.
34. De Pauw K, Roelands B, Cheung SS, *et al.* (2013) Guidelines to classify subject groups in sport-science research. *Int J Sports Physiol Perform* **8**, 111–122.
35. JPT Higgins & Green S (editors) (2011) *Cochrane Handbook for Systematic Reviews of Interventions*, version 5.1.0 (updated March 2011). The Cochrane Collaboration. http://handbook.cochrane.org
36. Cohen J (1988) *Statistical Power Analysis for the Behavioral Sciences*, 2nd ed. Hillsdale, NJ: Lawrence Earlbaum Associates.
37. Sterne JA, Sutton AJ, Ioannidis JP, *et al.* (2011) Recommendations for examining and interpreting funnel plot asymmetry in meta-analyses of randomised controlled trials. *BMJ* **343**, d4002.
38. Aucouturier J, Boissiere J, Pawlak-Chaouch M, *et al.* (2015) Effect of dietary nitrate supplementation on tolerance to supramaximal intensity intermittent exercise. *Nitric Oxide* **49**, 16–25.
39. Bailey SJ, Varnham RL, DiMenna FJ, *et al.* (2015) Inorganic nitrate supplementation improves muscle oxygenation, O_2 uptake kinetics, and exercise tolerance at high but not low pedal rates. *J Appl Physiol* **118**, 1396–1405.
40. Breese BC, McNarry MA, Marwood S, *et al.* (2013) Beetroot juice supplementation speeds O_2 uptake kinetics and improves exercise tolerance during severe-intensity exercise initiated from an elevated metabolic rate. *Am J Physiol Regul Integr Comp Physiol* **305**, R1441–R1450.
41. Buck CL, Henry T, Guelfi K, *et al.* (2015) Effects of sodium phosphate and beetroot juice supplementation on repeated-sprint ability in females. *Eur J Appl Physiol* **115**, 2205–2213.
42. Christensen PM, Petersen NK, Friis SN, *et al.* (2017) Effects of nitrate supplementation in trained and untrained muscle are modest with initial high plasma nitrite levels. *Scand J Med Sci Sports* **27**, 1616–1626.
43. Coggan AR, Leibowitz JL, Kadkhodayan A, *et al.* (2015) Effect of acute dietary nitrate intake on maximal knee extensor speed and power in healthy men and women. *Nitric Oxide* **48**, 16–21.
44. Corry LR & Gee TI (2015) Dietary nitrate enhances power output during the early phases of maximal intensity sprint cycling. *Int J Coaching Sci* **9**, 87–97.
45. Fulford J, Winyard PG, Vanhatalo A, *et al.* (2013) Influence of dietary nitrate supplementation on human skeletal muscle metabolism and force production during maximum voluntary contractions. *Pflugers Arch* **465**, 517–528.
46. Kelly J, Vanhatalo A, Wilkerson DP, *et al.* (2013) Effects of nitrate on the power-duration relationship for severe-intensity exercise. *Med Sci Sports Exerc* **45**, 1798–1806.
47. Kokkinoplitis K & Chester N (2014) The effect of beetroot juice on repeated sprint performance and muscle force production. *J Phys Educ Sport* **14**, 242–247.
48. Mosher SL, Sparks SA, Williams EL, *et al.* (2016) Ingestion of a nitric oxide enhancing supplement improves resistance exercise performance. *J Strength Cond Res* **30**, 3520–3524.
49. Murphy M, Eliot K, Heuertz RM, *et al.* (2012) Whole beetroot consumption acutely improves running performance. *J Acad Nutr Diet* **112**, 548–552.
50. Nyakayiru J, Jonvik KL, Trommelen J, *et al.* (2017) Beetroot juice supplementation improves high-intensity intermittent type exercise performance in trained soccer players. *Nutrients* **9**, E314.
51. Porcelli S, Ramaglia M, Bellistri G, *et al.* (2015) Aerobic fitness affects the exercise performance responses to nitrate supplementation. *Med Sci Sports Exerc* **47**, 1643–1651.
52. Rienks JN, Vanderwoude AA, Maas E, *et al.* (2015) Effect of beetroot juice on moderate-intensity exercise at a constant rating of perceived exertion. *Int J Exerc Sci* **8**, 277–286.
53. Thompson KG, Turner L, Prichard J, *et al.* (2014) Influence of dietary nitrate supplementation on physiological and cognitive responses to incremental cycle exercise. *Respir Physiol Neurobiol* **193**, 11–20.
54. Thompson C, Wylie LJ, Fulford J, *et al.* (2015) Dietary nitrate improves sprint performance and cognitive function during prolonged intermittent exercise. *Eur J Appl Physiol* **115**, 1825–1834.
55. Vasconcellos J, Henrique Silvestre D, Dos Santos Baiao D, *et al.* (2017) A single dose of beetroot gel rich in nitrate does not improve performance but lowers blood glucose in physically active individuals. *J Nutr Metabol* **2017**, 7853034.
56. Wylie LJ, Mohr M, Krustrup P, *et al.* (2013) Dietary nitrate supplementation improves team sport-specific intense intermittent exercise performance. *Eur J Appl Physiol* **113**, 1673–1684.
57. Wylie LJ, Bailey SJ, Kelly J, *et al.* (2016) Influence of beetroot juice supplementation on intermittent exercise performance. *Eur J Appl Physiol* **116**, 415–425.
58. Boorsma RK, Whitfield J & Spriet LL (2014) Beetroot juice supplementation does not improve performance of elite 1500-m runners. *Med Sci Sports Exerc* **46**, 2326–2334.
59. Callahan MJ, Parr EB, Hawley JA, *et al.* (2017) Single and combined effects of beetroot crystals and sodium bicarbonate on 4-km cycling time trial performance. *Int J Sport Nutr Exerc Metab* **27**, 271–278.
60. Glaister M, Pattison JR, Muniz-Pumares D, *et al.* (2015) Effects of dietary nitrate, caffeine, and their combination on 20-km cycling time trial performance. *J Strength Cond Res* **29**, 165–174.
61. Hoon MW, Hopkins WG, Jones AM, *et al.* (2014) Nitrate supplementation and high-intensity performance in competitive cyclists. *Appl Physiol Nutr Metab* **39**, 1043–1049.
62. Hoon MW, Jones AM, Johnson NA, *et al.* (2014) The effect of variable doses of inorganic nitrate-rich beetroot juice on simulated 2000-m rowing performance in trained athletes. *Int J Sport Physiol* **9**, 615–620.
63. Kramer SJ, Baur DA, Spicer MT, *et al.* (2016) The effect of six days of dietary nitrate supplementation on performance in trained CrossFit athletes. *J Int Soc Sports Nutr* **13**, 39.
64. Lane SC, Hawley JA, Desbrow B, *et al.* (2014) Single and combined effects of beetroot juice and caffeine supplementation on cycling time trial performance. *Appl Physiol Nutr Metab* **39**, 1050–1057.

British Journal of Nutrition

65. Lowings S, Shannon OM, Deighton K, *et al.* (2017) Effect of dietary nitrate supplementation on swimming performance in trained swimmers. *Int J Sport Nutr Exerc Metab* **27**, 377–384.
66. Martin K, Smee D, Thompson KG, *et al.* (2014) No improvement of repeated-sprint performance with dietary nitrate. *Int J Sports Physiol Perform* **9**, 845–850.
67. McQuillan JA, Dulson DK, Laursen PB, *et al.* (2017) Dietary nitrate fails to improve 1 and 4 km cycling performance in highly trained cyclists. *Int J Sport Nutr Exerc Metab* **27**, 255–263.
68. McQuillan JA, Dulson DK, Laursen PB, *et al.* (2017) The effect of dietary nitrate supplementation on physiology and performance in trained cyclists. *Int J Sports Physiol Perform* **12**, 684–689.
69. Muggeridge DJ, Howe CC, Spendiff O, *et al.* (2013) The effects of a single dose of concentrated beetroot juice on performance in trained flatwater kayakers. *Int J Sport Nutr Exerc Metab* **23**, 498–506.
70. Nyakayiru JM, Jonvik KL, Pinckaers PJ, *et al.* (2017) No effect of acute and 6-day nitrate supplementation on VO_2 and time-trial performance in highly trained cyclists. *Int J Sport Nutr Exerc Metab* **27**, 11–17.
71. Peeling P, Cox GR, Bullock N, *et al.* (2015) Beetroot juice improves on-water 500 M time-trial performance, and laboratory-based paddling economy in national and international-level Kayak athletes. *Int J Sport Nutr Exerc Metab* **25**, 278–284.
72. Rimer EG, Peterson LR, Coggan AR, *et al.* (2016) Acute dietary nitrate supplementation increases maximal cycling power in athletes. *Int J Sports Physiol Perform* **11**, 715–720.
73. Rimer EG, Peterson LR, Coggan AR, *et al.* (2016) Increase in maximal cycling power with acute dietary nitrate supplementation. *Int J Sports Physiol Perform* **11**, 715–720.
74. Shannon OM, Barlow MJ, Duckworth L, *et al.* (2017) Dietary nitrate supplementation enhances short but not longer duration running time-trial performance. *Eur J Appl Physiol* **117**, 775–785.
75. Thompson C, Vanhatalo A, Jell H, *et al.* (2016) Dietary nitrate supplementation improves sprint and high-intensity intermittent running performance. *Nitric Oxide* **61**, 55–61.
76. Wilkerson DP, Hayward GM, Bailey SJ, *et al.* (2012) Influence of acute dietary nitrate supplementation on 50 mile time trial performance in well-trained cyclists. *Eur J Appl Physiol* **112**, 4127–4134.
77. Goulet ED (2011) Effect of exercise-induced dehydration on time-trial exercise performance: a meta-analysis. *Br J Sports Med* **45**, 1149–1156.
78. Larsen FJ, Schiffer TA, Borniquel S, *et al.* (2011) Dietary inorganic nitrate improves mitochondrial efficiency in humans. *Cell Metab* **13**, 149–159.
79. Hernández A, Schiffer TA, Ivarsson N, *et al.* (2012) Dietary nitrate increases tetanic $[Ca^{2+}]i$ and contractile force in mouse fast-twitch muscle. *J Physiol* **590**, 3575–3583.
80. Ferguson SK, Hirai DM, Copp SW, *et al.* (2013) Impact of dietary nitrate supplementation via beetroot juice on exercising muscle vascular control in rats. *J Physiol* **591**, 547–557.
81. McNamara TC, Keen JT, Simmons GH, *et al.* (2014) Endothelial nitric oxide synthase mediates the nitric oxide component of reflex cutaneous vasodilatation during dynamic exercise in humans. *J Physiol* **592**, 5317–5326.
82. Larsen FJ, Ekblom B, Sahlin K, *et al.* (2006) Effects of dietary nitrate on blood pressure in healthy volunteers. *New Eng J Med* **355**, 2792–2793.
83. McConell GK, Bradley SJ, Stephens TJ, *et al.* (2007) Skeletal muscle nNOS mu protein content is increased by exercise training in humans. *Am J Physiol Regul Integr Comp Physiol* **293**, R821–R828.
84. Tesch PA & Karlsson J (1985) Muscle fiber types and size in trained and untrained muscles of elite athletes. *J Appl Physiol (1985)* **59**, 1716–1720.
85. Currell K & Jeukendrup AE (2008) Validity, reliability and sensitivity of measures of sporting performance. *Sports Med* **38**, 297–316.
86. Amann M, Hopkins WG & Marcora SM (2008) Similar sensitivity of time to exhaustion and time-trial time to changes in endurance. *Med Sci Sports Exerc* **40**, 574–578.
87. Jeukendrup A, Saris WH, Brouns F, *et al.* (1996) A new validated endurance performance test. *Med Sci Sports Exerc* **28**, 266–270.
88. Jeukendrup AE & Currell K (2005) Should time trial performance be predicted from three serial time-to-exhaustion tests? *Med Sci Sports Exerc* **37**, 1820 author reply 1821.
89. Alexander J, Diane B, Cockburn A, *et al.* (2008) Nitrate in vegetables scientific opinion of the Panel on Contaminants in the Food chain. *EFSA J* **689**, 1–79.

British Journal of Nutrition