Effects of caffeine on time-trial performance and associated physiological responses: a meta-analysis

Mark Glaister[1], Gavin Moir[2]

[1]School of Sport, Health, and Applied Sciences, St Mary's University, Strawberry Hill, Twickenham, UK. [2]Department of Exercise Science, East Stroudsburg University, East Stroudsburg, PA, USA.

Corresponding Author:

Dr Mark Glaister

School of Sport, Health, and Applied Sciences

St. Mary's University

Waldegrave Road

Strawberry Hill

Twickenham

UK

TW1 4SX

Tel: (+44)208 240 4012

Fax: (+44)208 240 4212

E-mail: mark.glaister@stmarys.ac.uk

Running title: Caffeine, time trial, and physiology

Key words: Ergogenic aids, methylxanthine, endurance exercise, adenosine receptor, power output.

Authorship Confirmation Statement

MG wrote the introduction and performed the literature search. Both authors checked the literature for relevant papers. MG extracted the data on all the key variables and GM checked those data for accuracy. GM conducted the review of research quality of included articles and MG checked that review. GM converted all the performance data to power outputs and wrote the corresponding sections in the paper. MG conducted the meta-analyses on the key variables and wrote the corresponding methods and results sections. MG wrote the discussion section of the review. All authors have reviewed and approved of the manuscript prior to submission. We can confirm that the manuscript has been submitted solely to this journal and is not published, in press, or submitted elsewhere.

Abstract

Background: The aim was to conduct a systematic review and meta-analysis on the effects of caffeine supplementation on time-trial performance and associated physiological responses. **Methods**: 35 studies met the inclusion criteria of adopting double-blind, randomized, placebo-controlled, crossover designs that included a closed-loop time-trial (≥ 5 mins) performed under a caffeine dose of 3 – 6 $mg \cdot kg^{-1}$ administered 30 – 90 minutes beforehand. Meta-analyses were completed using a random-effects model, with effects on time-trial performance presented as standardized mean difference (δ) and with physiological responses presented as raw mean difference (*D*). 95% confidence limits (CL_{95}) were calculated for all estimates. **Results**: Relative to placebo, caffeine had a positive effect on time-trial performance (δ = .32; CL_{95} [.19, .44]). Moreover, the effect of caffeine on time-trial performance corresponded with increases in heart rate (*D* = 3.3 $b \cdot min^{-1}$; CL_{95} [1.7, 4.8]), oxygen uptake (*D* = .09 $L \cdot min^{-1}$; CL_{95} [.02, .17]), blood lactate (*D* = 1.42 $mmol \cdot L^{-1}$; CL_{95} [1.09, 1.74]), and blood glucose (*D* = .94 $mmol \cdot L^{-1}$; CL_{95} [.58, 1.30]). In contrast, caffeine had no effect on time-trial measures of respiratory exchange ratio (*D* = .01; CL_{95} [-.01, .02]), or ratings of perceived exertion (*D* = .1; CL_{95} [-.1, .3]). **Conclusion**: The results reveal a clear effect of caffeine on moderate to high-intensity time-trial performance. When considered in conjunction with research using fixed-intensity exercise, the caffeine-induced increase in time-trial intensity likely explains all of the associated increases in heart rate and oxygen uptake, and part of the increase in blood lactate and blood glucose.

Introduction

Caffeine is a socially acceptable drug consumed worldwide by over 90% of adults with no apparent long-term adverse health effects.[1] The ergogenic benefits of caffeine have been observed most consistently during moderate- to high-intensity aerobic exercise, with doses of 3 – 6 $mg \cdot kg^{-1}$ ingested 30 – 90 minutes prior to exercise leading to improvements of 1 – 6% in closed-loop time-trials.[1,2] The mechanism by which caffeine exerts its ergogenic effect most likely resides in the ability of caffeine to act as an adenosine receptor antagonist.[3] Nevertheless, the ubiquitous nature of adenosine receptors, coupled with their ability to activate and inhibit the same signalling cascades[4,5] has made it difficult to confirm the precise mechanism(s) by which caffeine exerts its ergogenic effect.

One of the main difficulties when trying to establish the effects of caffeine on physiological responses to exercise is distinguishing the direct effects of caffeine on physiological responses from those associated with the corresponding increase in time-trial intensity. A recent meta-analysis showed that during fixed-intensity exercise, typical of that experienced during moderate- to high-intensity aerobic exercise (60-85% $\dot{V}O_{2max}$), caffeine led to significant increases in minute ventilation, blood lactate concentration ([BLa]), and blood glucose concentration ([BGl]); as well having a suppressive effect on ratings of perceived exertion (RPE).[6] In contrast, caffeine had no significant effects on heart rate, respiratory exchange ratio (RER), or oxygen uptake ($\dot{V}O_2$).[6] Establishing the effects of caffeine on those same physiological responses during time-trial performance should help to clarify how each is affected by the corresponding increase in exercise intensity. The aim of this study was therefore to carry out a systematic review and meta-analysis of the effects of caffeine supplementation on moderate- to high-intensity closed-loop time-trial performance and associated physiological responses.

Methods

Systematic review

The databases of Pubmed, SportDiscus, Science Direct, and Web of Science were searched for peer-reviewed publications (prior to January 2018) containing 'caffeine' in the title or the abstract, along with the words 'endurance' or 'time-trial', but not 'to exhaustion'. Reference lists of those studies that passed the initial screening for potential inclusion in the analysis along with those from relevant review articles[2,3,7-13] and textbooks[1] were also examined for publications which may have eluded the search of online databases.

Inclusion and exclusion criteria

Studies considered for inclusion in this investigation were limited to those conducted on adult (age: ≥ 18 years) humans, which had adopted double-blind, randomized, placebo-controlled, crossover designs using a standard effective caffeine dose of 3 – 6 $mg \cdot kg^{-1}$ administered 30 – 90 minutes prior to exercise. The choice of dose was based on evidence that the effects of caffeine on endurance performance follow an inverted-U response pattern, with optimal responses within the 3 – 6 $mg \cdot kg^{-1}$ range.[14] Indeed, few studies have examined the effects of low caffeine doses (< 3 $mg \cdot kg^{-1}$) on endurance performance, and those that have report conflicting results.[14] Similarly, the decision to restrict the timing of caffeine administration to 30 – 90 minutes before exercise was due to the fact that peak plasma caffeine concentrations are reported to occur within this time period[1] and as such, this is the most common administration strategy. Studies examining combinations of supplements were included in the analysis if the experimental design incorporated a 'caffeine only' versus placebo comparison.[15-20] In cases where studies had investigated the effects of different caffeine doses,[21-25] the dose

closest to the upper limit of the inclusion range was used in the analysis. Moderate- to high-intensity closed-loop time-trials were defined as bouts of exercise lasting ≥ 5 mins during which participants were required to complete either: a) a set distance in the fastest time possible; b) a fixed amount of work in the fastest time possible; or c) as much work as possible in a prescribed time. Studies which included bouts of exercise, other than for warm-up or submaximal physiological assessment purposes, prior to the time-trial were excluded from the analysis due to the potential confounding influence of fatigue on subsequent time-trial performance. Studies investigating the influence of caffeine on time-trial performance in extreme environmental conditions were also excluded from the analysis. Research quality was evaluated by means of the Physiotherapy Evidence Database (PEDro) scale, which ranks the quality of research, via a series of questions, on a 10-point scale.[26] In line with the meta-analysis by Ganio et al.,[2] publications achieving a score < 6 were considered to lack sufficient quality to be included in the meta-analysis. No inclusion restrictions were placed on potential moderator variables of gender, training status, caffeine habituation, or supplementation method, since previous research has been unable to confirm whether any of those variables influence the effects of caffeine on endurance performance.[1] However, subgroup meta-analyses were used to investigate potential influences of supplementation method, dosage, and exercise duration on time-trial performance and associated physiological responses to caffeine (see below).

Data extraction

For the meta-analysis, data were extracted from relevant publications as means, standard deviations (SD), and sample sizes. In instances where data were presented in a graphical format, images were enlarged to improve the precision of the data estimates. For the rare occasions where data was missing, authors were contacted to try to resolve the issue.

Physiological responses were limited to those which were most commonly evaluated during time-trials, which were: mean heart rate, mean $\dot{V}O_2$, mean RER, end-test RPE, end-test [BLa], and end-test [BGl]. Measures of RPE were constrained to those evaluated using the 15-point scale.[28]

Meta-analysis

From an initial search result of 934 studies, 35 met the inclusion criteria for the meta-analysis (Table 1). Meta-analyses were conducted using specialist software (Review Manager Version 5.3. The Nordic Cochrane Centre, Copenhagen: The Cochrane Collaboration, 2014). Meta-analyses were completed using a random-effects model with time-trial responses presented as a standardized mean difference (δ) and with physiological responses presented as raw mean difference (*D*). 95% confidence limits (CL_{95}) were calculated for all estimates. Time-trial data are presented as mean power outputs to provide consistency in the format with which these data are reported and to provide a more meaningful interpretation of the data for the reader. In those instances where time-trial data were presented in a format other than power output, data were converted as follows:

Mean power output conversions for the cycling time trials were performed as outlined by Martin et al.[52]

$$Power\ output = \frac{\left((0.5CdA \times \rho \times \bar{v}^2) + (m \times Crr \times g)\right) \times \bar{v}^2}{(1 - DTloss/100)}$$

where CdA is the product of the drag coefficient and frontal area (a fixed value of 0.321 was used), ρ is air density (using a fixed value of 1.226 kg·m^{-3}), $\bar{v}$ is the average velocity, m is the total mass of rider and bicycle (a fixed value of 8 kg was used for the bicycle), Crr is the

coefficient of rolling resistance (using a fixed value of 0.005), g is the gravitational acceleration of 9.8067 m·s^{-2}, $DTloss$ is the drivetrain loss (using a fixed value of 3%). The time trials used by McNaughton et al.[43,44] included simulated hill climbs that resulted in power outputs that were substantially lower than those reported by other authors using time trials of similar distances with cyclists of similar physiological characteristics.[19,20,23,41,47] Therefore, mean power outputs for the placebo trials completed by McNaughton et al.[43,44] were predicted from those presented by others[19,20,23,41,47] using regression analysis. The power outputs for the caffeine trials completed by McNaughton et al.[43,44] were then calculated using the percentage increase reported in the original papers.

Mean power output during running trials were calculated using the following equation from Helene and Yamashita:[53]

$$Power\ output = m\frac{dv}{t}v + \frac{CdρA}{2}(v - v_{wind})^2 v + PO_{vert}$$

where *m* is the mass of the subjects, *v* is the average running speed, *Cd* is the drag coefficient (a fixed value of 0.5 was used), ρ is air density (using a fixed value of 1.2 kg·m^{-3}), *A* is the frontal area (a fixed value of 1 m^2 was used), v_{wind} is the tailwind speed (0 m·s^{-1}), PO_{vert} is the power expended due to vertical motion of the center of mass. PO_{vert} was calculated using the vertical motion of the center of mass predicted from the regression equation developed by Lee and Farley[54] and the step frequencies presented by de Ruiter et al.[55] with a value of 2.81 Hz used for experienced runners[16,34] and a value of 2.59 Hz used for novice runners.[35]

Power output was estimated from time-trial speed during rowing tasks using the following equation provided by the manufacturer of the Concept II rowing ergometer:

$$Power\ output = 2.8 \times \left(\frac{d}{t}\right)^3$$

where d is the distance of the time trial and t is completion time.

The mean power outputs for time trial tasks performed on skiing ergometers were converted using the energy cost of 0.70 $J \cdot m^{-1} \cdot kg^{-1}$ reported by Pellegrini et al.[56] for the double-poling technique. The data from Berglund and Hemmingson[30] were subjected to the same conversion. However, as those authors did not report body mass for the subjects, an average body mass of 66.8 kg was determined for a mixed group of male and female cross-country skiers based upon previous research.[48,49,57]

For the hand cycling study,[39] the paper by Conger and Bassett[58] was used to predict metabolic energy expenditure from speed using linear regression. Power output was then estimated based on an assumed gross mechanical efficiency of 12.1% from previous research.[59]

Heterogeneity between studies was examined using the I^2 statistic, which describes the percentage of variability in mean difference estimates due to heterogeneity rather than chance. When I^2 was > 25% (25 – 50% represents moderate heterogeneity[60]), a subgroup meta-analysis was completed to investigate the source of heterogeneity. In line with recommendations regarding tests for heterogeneity,[61] CL_{95} for I^2 were calculated using the method outlined by Higgins & Thompson.[62] Subgroup meta-analyses were performed, when appropriate, to investigate the influence of the following potential moderator variables: 1) exercise duration, which was evaluated using meta-regression (Comprehensive Meta-analysis software Version 2.2; Biostat Inc., Englewood, NJ); 2) supplementation method (capsule versus drink formats);

and 3) caffeine dose (constrained to comparisons between the upper [≥ 5 mg·kg^{-1}] and lower [< 5 mg·kg^{-1}] half of the inclusion range). Of the remaining potential moderator variables, no comparisons were made to investigate the effects of: 1) exercise mode: since most had used cycling (n = 22) and there was no rationale to expect any differential effects of exercise mode on the response to caffeine; 2) gender: since only five studies[18,19,21,29,50] had used solely female participants; 3) training status: since between-study inconsistences in the way that this variable was reported/measured did not allow quantification with adequate precision; and 4) administration time: since most studies had administered the supplement 60 minutes prior to exercise (n = 25). Heterogeneity between subgroups was also evaluated using the I^2 statistic. Statistical significance was accepted at $p < 0.05$ for all analyses.

Results

Time trial

Relative to placebo, caffeine led to a significant increase (4.4 ± 3.1%) in time-trial performance (Figure 1) (δ = .32; CL_{95} [.19, .44]; $p < .00001$; n = 532), which translated into an increase in mean power output of 10.0 ± 8.4 W. There was no evidence of heterogeneity between the 41 studies that were included in the analysis (I^2 = 0%; CL_{95}[0, 37]).

Heart rate and perceived exertion

The effects of caffeine on heart rate and RPE during the time-trials are presented in Figure 2. Relative to placebo, caffeine supplementation resulted in a significant increase in heart rate (D = 3.3 b·min^{-1}; CL_{95}[1.7, 4.8]; $p < .0001$; n = 227) but had no effect on RPE (D = .1 [-.1, .3]; p = .47; n = 212). There was no evidence of heterogeneity between the studies evaluating heart rate (I^2 = 0%; CL_{95}[0, 49]), or RPE (I^2 = 0%; CL_{95}[0, 52]).

Oxygen uptake and respiratory exchange ratio

In comparison with placebo, caffeine resulted in a significant increase in $\dot{V}O_2$ (D =.09 L·min^{-1}; CL_{95}[.02, .17]; p = .02; n = 143) during the time trials but had no effect on RER (D = .01; CL_{95}[-.01, .02]; p = .32; n = 125) (Figure 3). Although there was no evidence of heterogeneity between the studies that evaluated $\dot{V}O_2$ (I^2 = 0%; CL_{95}[0, 60]), there was evidence of moderate heterogeneity between the studies that analysed RER (I^2 = 30%; CL_{95}[0, 68]). Subgroup analysis of the studies that evaluated RER was difficult given that only one study had used a caffeine dose < 5 mg·kg^{-1} or had administered caffeine in drink format (Table 2). Nevertheless, the lack of a significant effect of caffeine on RER remained regardless of subgroup and there was no evidence of heterogeneity between the subgroups. Moreover, meta-regression analysis revealed no relationship (r = .0001 [-.0005, .0007]) between exercise duration and the effect of caffeine, relative to placebo, on RER (Figure 4).

Blood lactate and blood glucose

The effects of caffeine on [BGl] and [BLa] during the time-trials are presented in Figure 5. Relative to placebo, caffeine resulted in significant increases in [BGl] (D = .94 mmol·L^{-1} [.58, 1.30]; p < .00001; n = 105) and [BLa] (D = 1.42 mmol·L^{-1} [1.09, 1.74]; p < .00001; n = 222). There was no evidence of heterogeneity between the studies evaluating [BGl] (I^2 = 0%; CL_{95}[0, 62]), or [BLa] (I^2 = 0%; CL_{95}[0, 51]).

Discussion

The aim of this study was to conduct a systematic review and meta-analysis on the effects of caffeine supplementation on closed-loop time-trial performance and associated physiological

responses. The main findings were that, relative to placebo, caffeine supplementation resulted in significant increases in time-trial performance and corresponding increases in heart rate, $\dot{V}O_2$, [BGl] and [BLa]. In contrast, caffeine had no effect on time-trial measures of RER or RPE.

The increase in time-trial performance confirms previous reports that a 3 – 6 $mg \cdot kg^{-1}$ caffeine dose administered approximately 60 minutes prior to exercise leads to an increase in time-trial performance of 1-6%.[1,2] Moreover, the response does not appear to be influenced by the size of the dose within that range, the method of administration, or the duration of exercise (at least when the duration is ≥ 5 minutes). Indeed, the absence of between-study heterogeneity suggests also that the effect of caffeine on time-trial performance is consistent irrespective of differences in exercise mode, training status, or gender. The mechanisms to explain the effects of caffeine on time-trial performance are difficult to elucidate but appear to be due most likely to the ability of caffeine to act as an adenosine receptor antagonist,[3] thereby influencing glucose homeostasis and lipid metabolism,[4] central nervous system function,[63] and cardiovascular and respiratory responses.[64] Nevertheless, the fact that adenosine receptors have four subtypes (A_1, A_{2A}, A_{2B}, and A_3) with the ability to activate and inhibit the same signalling cascades[4,5] makes it difficult to identify the precise mechanisms by which caffeine exerts its effects.

Some studies have suggested that caffeine may also influence performance via a direct effect on intracellular calcium mobilisation, at least during submaximal exercise.[12] However, effects via that mechanism are still unclear.[3] There is also some evidence that the effect of caffeine on time-trial performance may be influenced by a genetic factor.[27,34,35,51,65-67] Research to date has focused on the CYP1A2 gene which influences the rate at which the liver metabolises caffeine,

and the ADORA2A gene which, via its influence on A2A receptor binding characteristics, influences dopaminergic neurotransmission.[65,67] Results so far have been equivocal, possibly due to methodological inconsistencies.[65,67] Moreover, given that the ergogenic effects of caffeine happen in advance its metabolism, and during both long and relatively short time-trials, it is difficult to reconcile the role of the CYP1A2 gene in the ergogenic effect of caffeine. Indeed, recent reviews into the role of genetics on the ergogenic effects of caffeine have highlighted that more work in the area is warranted; including replication of previous studies and an expansion of the number of biologically plausible genes.[65,67]

The increase in time-trial intensity resulting from caffeine supplementation provides the most likely explanation for the corresponding increases in heart rate and $\dot{V}O_2$; particularly given that during fixed–intensity exercise at 60 – 85% $\dot{V}O_{2max}$, caffeine is reported to have no effect on either response.[6] Indeed, the increases in heart rate and $\dot{V}O_2$ following caffeine supplementation are in-line with what would be expected typically given the magnitude of the corresponding increase in mean power output.[68,69] In contrast, the increases in [BGl] and [BLa] are approximately double the values observed during fixed-intensity (60 – 85% $\dot{V}O_{2max}$) exercise following caffeine supplementation,[6] suggesting that the caffeine-induced increase in time-trial intensity provides only part of the explanation for those responses.

During exercise [BGl] is maintained at a normal value of 4.0 – 5.5 $mmol \cdot L^{-1}$ by various physiological processes to ensure that hepatic glucose output matches cellular uptake.[70] As exercise intensity increases above the lactate threshold, the rate of hepatic glucose release (via glycogenolysis and gluconeogenesis) exceeds that of peripheral glucose uptake, resulting in an increase in [BGl].[70,71] Although the increase in [BGl] is transient when exercise is prolonged,[70]

it is important, at this stage, to recognise that participants tend to increase power output at the end of time-trials as the finishing point approaches,[16-18,31,32,42,45,47,48] leading to somewhat elevated end-test measures of [BGl], [BLa] and RPE. When exercise intensity is fixed, caffeine increases [BGl] relative to placebo by ~ 0.4 $mmol \cdot L^{-1}$, independent of exercise intensity,[6] and most likely via an impairment of peripheral glucose uptake.[6] Under the same conditions, caffeine increases [BLa] by ~ 0.7 $mmol \cdot L^{-1}$, though the mechanisms of the response are more difficult to resolve and cannot easily be explained by effects on production or clearance.[6] Nevertheless, the caffeine-induced increase in time-trial intensity provides the most likely explanation for the additional increases in [BGl] and [BLa] above what are expected typically during fixed-intensity exercise and the changes are in-line with the expected responses.[71]

In contrast to the above, the absence of any effect of caffeine on RER during the time trials is difficult to explain; particularly when considering that an increase in time-trial intensity would normally be expected to increase RER due to a corresponding change in substrate metabolism.[72] Moreover, the caffeine-induced increase in [BLa] would normally be expected also to increase RER, as a result of an increase in H^+ buffering.[72] One possible explanation for the absence of an effect of caffeine on RER during the time-trials could lie in the fact that during fixed-intensity exercise there is evidence of an interaction effect between caffeine and exercise intensity on RER, with values reducing, relative to placebo, as exercise intensity increases;[48,73] Although the mechanisms to explain that response are unclear,[6] it is possible that the absence of an effect of caffeine on RER during the time-trials is due to caffeine counteracting the increase in RER that would be expected following an increase in exercise intensity.

Finally, the absence of any effect of caffeine supplementation on RPE during the time-trials, despite the increase in time-trial intensity, confirms previous research[9] showing that caffeine has a suppressive effect on perceptual responses during exercise leading to a reduction in RPE during fixed-intensity exercise[6] or an increase in performance for the same RPE response, as in the present study.

Conclusion

When consumed in a dose of 3 – 6 $mg \cdot kg^{-1}$, 30 – 90 minutes prior to exercise lasting ≥ 5 minutes, there is a clear effect of caffeine on time-trial performance with no corresponding change in the perception of effort or substrate utilisation. Nevertheless, coaches and practitioners should be aware that those performance gains are likely to be accompanied by small corresponding increases in heart rate and $\dot{V}O_2$, and disproportionate increases in [BGl] and [BLa]. For researchers, the challenge is to identify the mechanisms by which caffeine improves time-trial performance and to establish the role, if any, of caffeine-induced increases in [BGl] and [BLa] on that response.

Acknowledgements

Many thanks to those authors who supplied us with missing data from their research.

Author Disclosure Statement

No competing financial interests exist.

References

1. Burke LM, Desbrow B, Spriet L. Caffeine for Sports Performance. Champaign (IL): Human Kinetics; 2013. p. 1-156.
2. Ganio MS, Klau JF, Casa DJ, et al. Effect of caffeine on sport-specific endurance performance: a systematic review. *J Strength Cond Res*. 2009;23(1):315-24.
3. Kalmar JM. The influence of caffeine on voluntary muscle activation. *Med Sci Sports Exerc*. 2005;37(12):2113-9.
4. Koupenova M, Ravid K. Adenosine, adenosine receptors and their role in glucose homeostasis and lipid metabolism. *J Cell Physiol*. 2013; doi:10.1002/jcp.24352.
5. Layland J, Carrick D, Lee M, et al. Adenosine: physiology, pharmacology, and clinical applications. *JACC: Cardiovasc Interv*. 2014;7(6):581-91.
6. Glaister M, Gissane C. Caffeine and physiological responses to submaximal exercise: a meta-analysis. *Int J Sports Physiol Perform*. 2018;13(4):402-11.
7. Graham TE. Caffeine and exercise. *Sports Med*. 2001;31(11):785-807.
8. Burke LM. Caffeine and sports performance. *Appl Physiol Nutr Metab*. 2008;33(6):1319-34.
9. Doherty M, Smith PM. Effects of caffeine ingestion on rating of perceived exertion during and after exercise: a meta-analysis. *Scand J Med Sci Sports*. 2005;15(2):69-78.
10. Goldstein ER, Ziegenfuss T, Kalman D, et al. International society of sports nutrition position stand: caffeine and performance. *J Int Soc Sports Nutr*. 2010;7(1):1-15.
11. Nehlig A, Debry G. Caffeine and sports activity: a review. *Int J Sports Med.* 1994;15(5):215-23.
12. Tarnopolsky MA. Effect of caffeine on the neuromuscular system-potential as an ergogenic aid. *Appl Physiol Nutr Metab*. 2008;33(6):1284-9.

13. Warren GL, Park, ND, Maresca RD, et al. Effect of caffeine ingestion on muscular strength and endurance: a meta-analysis. *Med Sci Sports Exerc*. 2010;42(7):1375-87.
14. Spriet LL. Exercise and sport performance with low doses of caffeine. *Sports Med*. 2014;44(2):S175-84.
15. Acker-Hewitt TL, Shafer BM, Saunders MJ, et al. Independent and combined effects of carbohydrate and caffeine ingestion on aerobic cycling performance in the fed state. *Appl Physiol Nutr Metab*. 2012;37(2):276-83.
16. Bell DG, McLellan TM, Sabiston CM. Effect of ingesting caffeine and ephedrine on 10-km run performance. *Med Sci Sports Exerc*. 2002;34(2):344-9.
17. Christensen PM, Petersen MH, Friis SN, et al. Caffeine, but not bicarbonate, improves 6 min maximal performance in elite rowers. *Appl Physiol Nutr Metab*. 2014;39(9):1058-63.
18. Glaister M, Pattison JR, Muniz-Pumares D, et al. Effects of dietary nitrate, caffeine, and their combination on 20-km cycling time trial performance. *J Strength Cond Res*. 2015;29(1):165-74.
19. Lane SC, Hawley JA, Desbrow B, et al. Single and combined effects of beetroot juice and caffeine supplementation on cycling time trial performance. *Appl Physiol Nutr Metab*. 2014;39(9):1050-7.
20. Quinlivan A, Irwin C, Grant GD, et al. The effects of Red Bull energy drink compared with caffeine on cycling time-trial performance. *Int J Sports Physiol Perform*. 2015;10(7):897-901.
21. Anderson ME, Bruce CR, Fraser SF, et al. Improved 2000-meter rowing performance in competitive oarswomen after caffeine ingestion. *Int J Sport Nutr Exerc Metab*. 2000;10(4):464-75.
22. Bruce CR, Anderson ME, Fraser SF, et al. Enhancement of 2000-m rowing performance after caffeine ingestion. *Med Sci Sports Exerc*. 2000;32(11):1958-63.

23. Desbrow B, Biddulph C, Devlin B, et al. The effects of different doses of caffeine on endurance cycling time trial performance. *J Sports Sci*. 2012;30(2):115-20.
24. Jenkins NT, Trilk JL, Singhal A, et al. Ergogenic effects of low doses of caffeine on cycling performance. *Int J Sport Nutr Exerc Metab*. 2008;18(3):328-42.
25. Skinner TL, Jenkins DG, Coombes JS, et al. Dose response of caffeine on 2000-m rowing performance. *Med Sci Sports Exerc*. 2010;42(3):571-6.
26. Verhagen AP, de Vet HC, de Bie RA, et al. The Delphi list: a criteria list for quality assessment of randomized clinical trials for conducting systematic reviews developed by Delphi consensus. *J Clin Epidemiol*. 1998;51(12):1235-41.
27. Algrain HA, Thomas RM, Carrillo AE, et al. The effects of a polymorphism in the cytochrome P450 CYP1A2 gene on performance enhancement with caffeine in recreational cyclists. *J Caffeine Res*. 2016;6(1):34-9
28. Borg G. Perceived exertion as an indicator of somatic stress. *Scand J Rehab Med*. 1970;2(2):92-8.
29. Astorino TA, Roupoli LR, Valdivieso BR. Caffeine does not alter RPE or pain perception during intense exercise in active women. *Appetite*. 2012;59(2):585-90.
30. Berglund B, Hemmingsson P. Effects of caffeine ingestion on exercise performance at low and high altitudes in cross-country skiers. *Int J Sports Med*. 1982;3(4):234-6.
31. Black CD, Waddell DE, Gonglach AR. Caffeine's ergogenic effects on cycling: neuromuscular and perceptual factors. *Med Sci Sports Exerc*. 2015;47(6):1145-58.
32. Bortolotti H, Altimari LR, Vitor-Costa M, et al. Performance during a 20-km cycling time-trial after caffeine ingestion. *J Int Soc Sports Nutr.* 2014;11(1):1-7.
33. Boyett JC, Giersch GE, Womack CJ, et al. Time of Day and Training Status Both Impact the Efficacy of Caffeine for Short Duration Cycling Performance. *Nutrients*. 2016;8(10):E639.

34. Bridge CA, Jones MA. The effect of caffeine ingestion on 8 km run performance in a field setting. *J Sports Sci*. 2006;24(4):433-9.
35. Church DD, Hoffman JR, LaMonica MB, et al. The effect of an acute ingestion of Turkish coffee on reaction time and time trial performance. *J Int Soc Sports Nutr*. 2015;12:37.
36. Felippe LC, Ferreira GA, Learsi SK, et al. Caffeine increases both total work performed above critical power and peripheral fatigue during a 4-km cycling time trial. *J Appl Physiol*. 2018; 124:1491–1501.
37. Giersch GEW, Boyett JC, Hargens TA, et al. The effect of the CYP1A2 - 163 C > A polymorphism on caffeine metabolism and subsequent cycling performance. *J Caffeine Adenosine Res*. 2018;8(2):65-70.
38. Gonçalves LS, Painelli VS, Yamaguchi G, et al. Dispelling the myth that habitual caffeine consumption influences the performance response to acute caffeine supplementation. *J Appl Physiol*. 2017;123(1):213-20.
39. Graham-Paulson T, Perret C, Goosey-Tolfrey V. Improvements in cycling but not handcycling 10 km time trial performance in habitual caffeine users. *Nutrients*. 2016;8(7):E393.
40. Hodgson AB, Randell RK, Jeukendrup AE. The metabolic and performance effects of caffeine compared to coffee during endurance exercise. *PLoS One*. 2013;8(4):e59561.
41. Irwin C, Desbrow B, Ellis A, et al. Caffeine withdrawal and high-intensity endurance cycling performance. *J Sports Sci*. 2011;29(5):509-15.
42. Laurence G, Wallman K, Guelfi KJ. Effects of caffeine on time trial performance in sedentary men. *J Sports Sci*. 2012;30(12):1235-40.
43. McNaughton LR, Lovell RJ, Siegler J, et al. The effects of caffeine ingestion on time trial cycling performance. *Int J Sports Physiol Perform*. 2008;3(2):157-63.

44. McNaughton LR, Lovell RJ, Siegler JC, et al. The effects of caffeine ingestion on time trial cycling performance. *J Sports Med Phys Fitness*. 2008;48(3):320-5.
45. Santos R de A, Kiss MA, Silva-Cavalcante MD, et al. Caffeine alters anaerobic distribution and pacing during a 4000-m cycling time trial. PLoS One. 2013;8(9):e75399.
46. Saunders B, de Oliveira LF, da Silva RP, et al. Placebo in sports nutrition: a proof-of-principle study involving caffeine supplementation. *Scand J Med Sci Sports*. 2017;27(11):1240-7.
47. Skinner TL, Jenkins DG, Taaffe DR, et al. Coinciding exercise with peak serum caffeine does not improve cycling performance. *J Sci Med Sport*. 2013;16(1):54-9.
48. Stadheim HK, Kvamme B, Olsen R, et al. Caffeine increases performance in cross-country double-poling time trial exercise. *Med Sci Sports Exerc*. 2013;45(11):2175-83.
49. Stadheim HK, Spencer M, Olsen R, et al. Caffeine and Performance over Consecutive Days of Simulated Competition. *Med Sci Sports Exerc*. 2014;46(9):1787-96.
50. Wallman KE, Goh JW, Guelfi KJ. Effects of caffeine on exercise performance in sedentary females. *J Sports Sci Med.* 2010;9(2):183-9.
51. Womack CJ, Saunders MJ, Bechtel MK, et al. The influence of a CYP1A2 polymorphism on the ergogenic effects of caffeine. *J Int Soc Sports Nutr*. 2012;9(1):7.
52. Martin JC, Miliken DL, Cobb JE, et al. Validation of a mathematical model for road cycling power. *J Biomech*. 1998;14(3):276-91.
53. Helene O, Yamashita MT. The force, power and energy of the 100 meter sprint. *Am J Physics*. 2010;78(3):307-9.
54. Lee CA, Farley CT. Determinants of the center of mass trajectory in human walking and running. *J Exp Biol*. 1998;201(21):2935-44.
55. de Ruiter CJ, Verdijk PWL, Werker W, et al. Stride frequency in relation to oxygen consumption in experienced and novice runners. *Eur J Sport Sci*, 2013;14(3):251-8.

56. Pellegrini B, Zoppirolli C, Bortolan L, et al. Gait models and mechanical energy in three cross-country skiing techniques. *J Exp Biol*. 2014;217(21):3910-8.
57. Sandbakk Ø, Ettema G, Holmberg H-C. Gender differences in endurance performance by elite cross-country skiers are influenced by the contribution from poling. *Scand J Med Sci Sports*. 2014; 24(1):28-33.
58. Conger SA, Bassett DR. A compendium of energy costs of physical activities for individuals who use manual wheelchairs. *Adapt Phys Activ Q*. 2011;28(4):310-25.
59. van der Woude LHV, Horstman A, Faas P, et al. Power output and metabolic cost of synchronous and asynchronous submaximal and peak level hand cycling on a motor driven treadmill in able-bodied male subjects. *Med Eng Phys*. 2008;30(5):574-80.
60. Higgins JP, Thompson SG, Deeks JJ, et al. Measuring inconsistency in meta-analyses. *BMJ*. 2003;327(7414):557-60.
61. Ioannidis JPA, Patsopoulos NA, Evangelou E. Uncertainty in heterogeneity estimates in meta-analyses. *BMJ*. 2007;335(7626):914-6.
62. Higgins JP, Thompson SG. Quantifying heterogeneity in a meta analysis. *Stat Med*. 2002;21(11):1539-58.
63. Benarroch EE. Adenosine and its receptors: multiple modulatory functions and potential therapeutic targets for neurologic disease. *Neurology*. 2008;70(3):231-6.
64. Biaggioni I, Olafsson B, Robertson RM, et al. Cardiovascular and respiratory effects of adenosine in conscious man: evidence for chemoreceptor activation. *Circ Res*. 1987;61(6):779-86.
65. Fulton JL, Dinas PC, Carrillo AE, et al. Impact of genetic variability on physiological responses to caffeine in humans: a systematic review. *Nutrients*. 2018;10:1373.
66. Guest N, Corey P, Vescovi J, et al. Caffeine, CYP1A2 genotype, and endurance performance in athletes. *Med Sci Sports Exerc*. 2018;50(8):1570-78.

67. Southward K, Rutherfurd-Markwick K, Badenhorst C, et al. The role of genetics in moderating the inter-individual differences in the ergogenicity of caffeine. *Nutrients* 2018;10:1352.
68. Grazzi G, Alfieri N, Borsetto C, et al. The power output/heart rate relationship in cycling: test standardization and repeatability. *Med Sci Sports Exerc*. 1999;31(10):1478-83.
69. Neder JA, Nery LE, Peres C, et al. Reference values for dynamic responses to incremental cycle ergometry in males and females aged 20 to 80. *Am J Respir Crit Care Med*. 2001;164(8):1481-6.
70. Suh SH, Paik IY, Jacobs K. Regulation of blood glucose homeostasis during prolonged exercise. *Mol Cells*. 2007;23(3):272-9.
71. Simões HG, Grubert Campbell CS, et al. Blood glucose responses in humans mirror lactate responses for individual anaerobic threshold and for lactate minimum in track tests. *Eur J Appl Physiol Occup Physiol*. 1999;80(1):34-40.
72. Goedecke JH, St Clair Gibson A, Grobler L, et al. Determinants of the variability in respiratory exchange ratio at rest and during exercise in trained athletes. *Am J Physiol Endocrinol Metab*. 2000;279(6):E1325-34.
73. Glaister M, Williams BH, Muniz-Pumares D, et al. The effects of caffeine supplementation on physiological responses to submaximal exercise in endurance-trained men. *PLoS One*. 2016;11(8):e0161375.

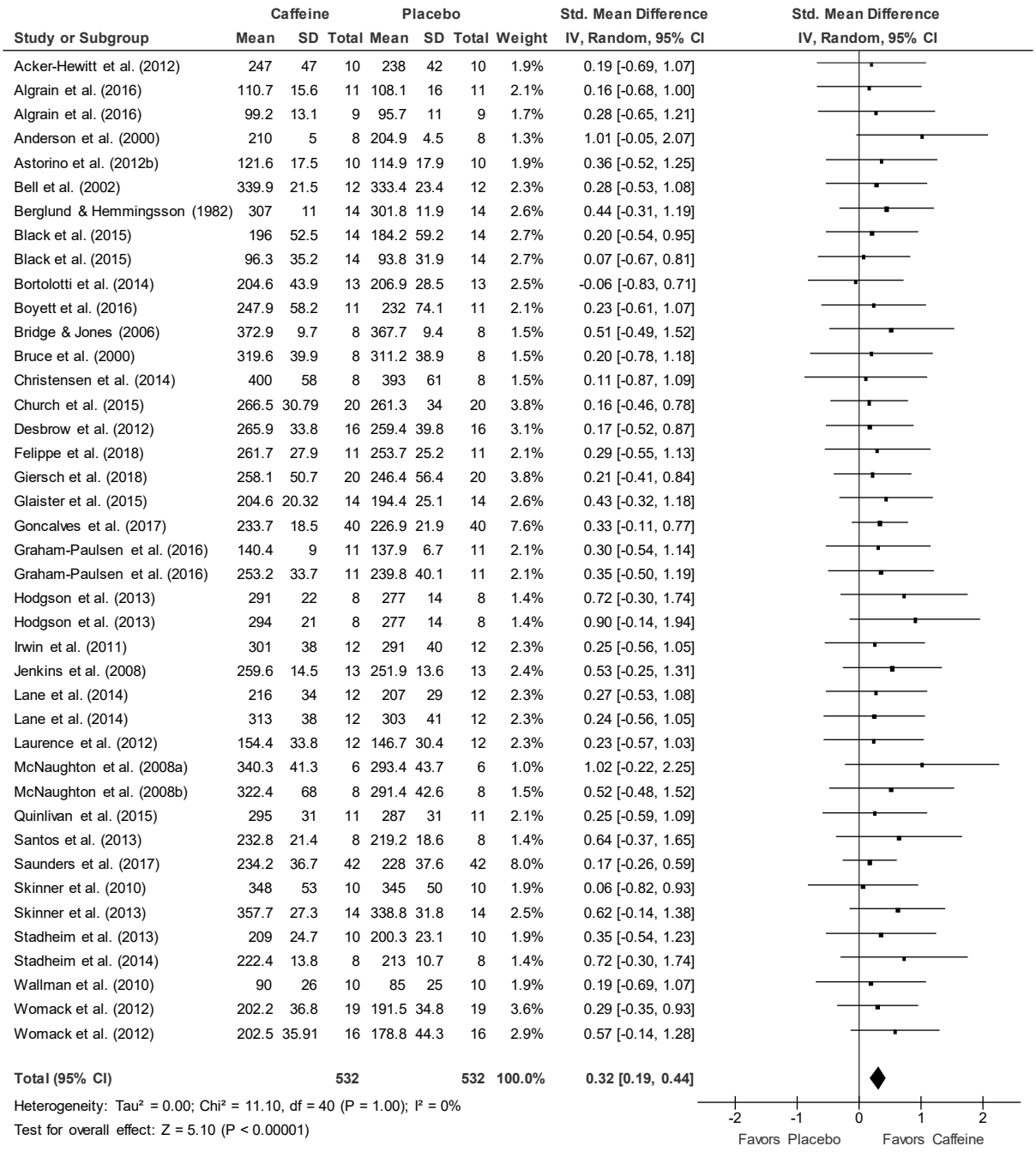
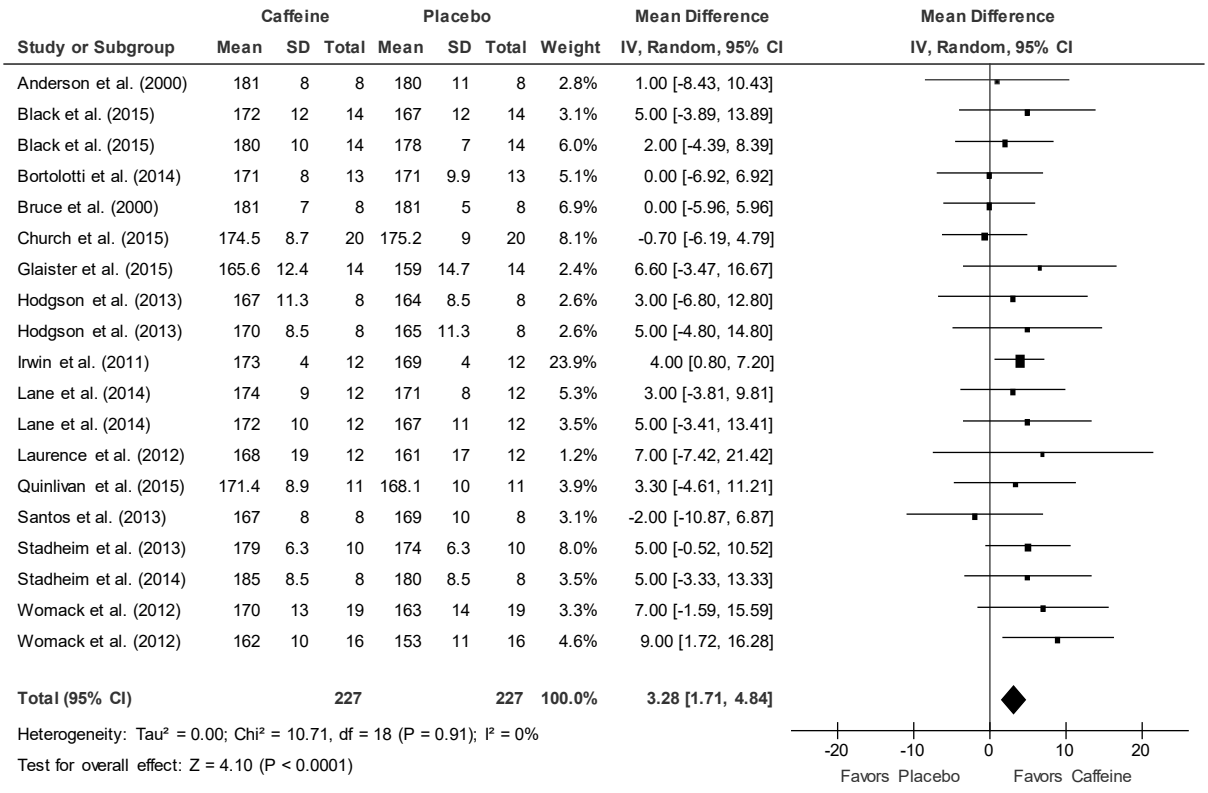


Figure 1. A forest plot of studies that have investigated the effects of caffeine supplementation on closed-loop time-trial (≥ 5 mins) performance. Squares represent the standardized mean difference, relative to placebo, with associated 95% confidence limits. The size of each square reflects the weighting given to each response. The diamond at the base of the plot represents the overall effect calculated from a random effects model; the width of the diamond representing the 95% confidence interval.



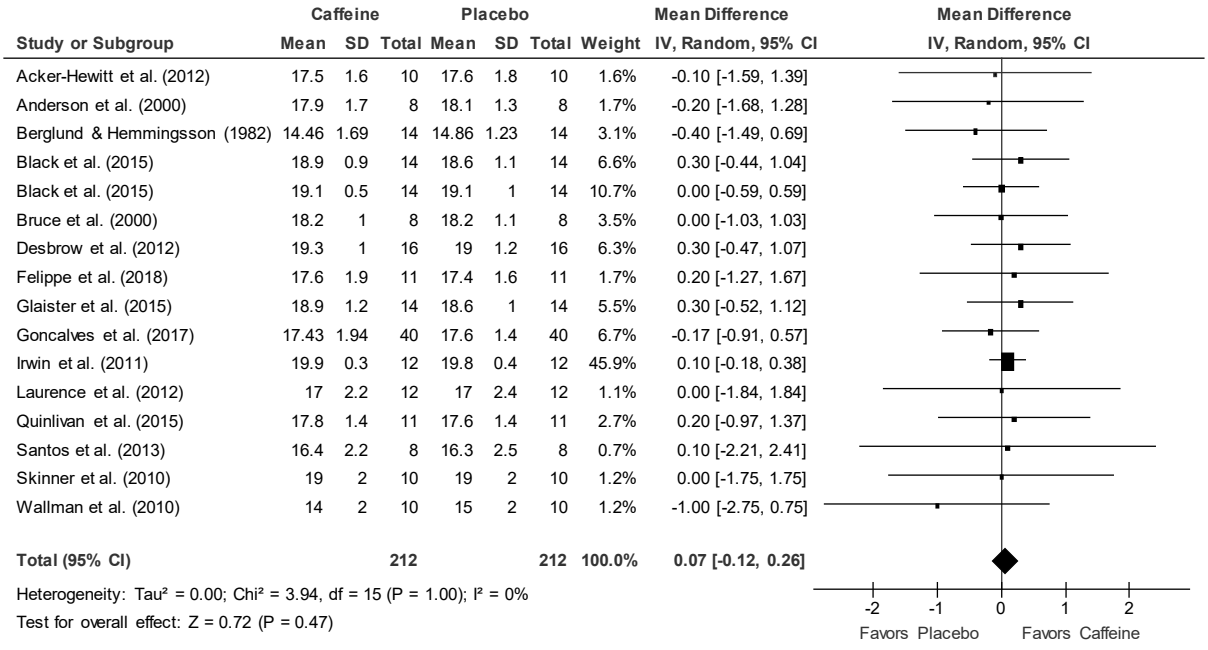
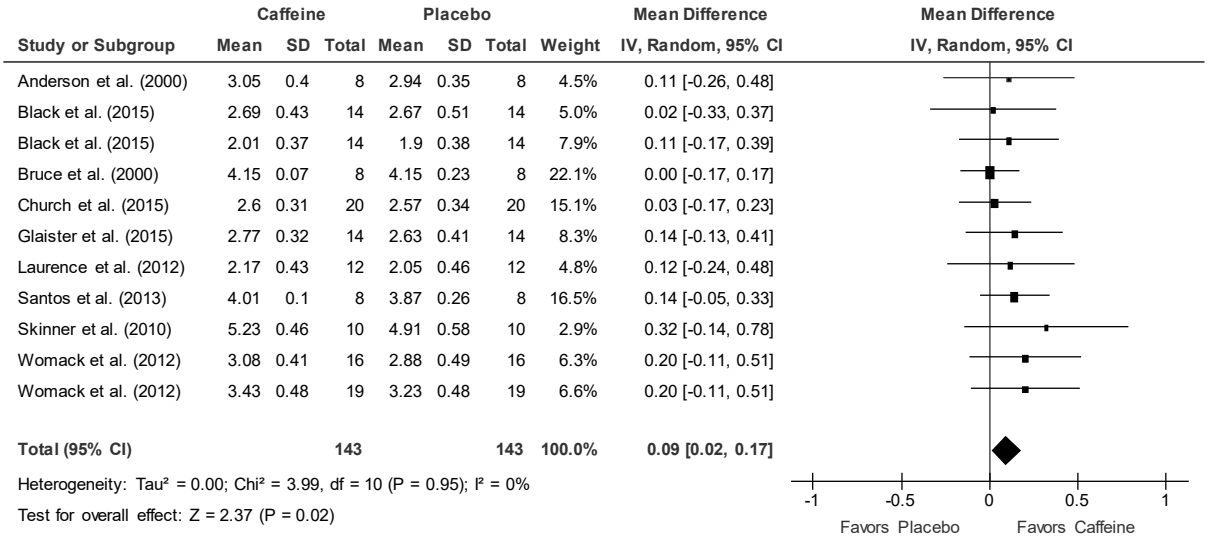


Figure 2. Forest plots of studies that have investigated the effects of caffeine supplementation on heart rate (upper plot), and ratings of perceived exertion (lower plot) during closed-loop time-trial (≥ 5 mins) performance. Squares represent the raw mean difference, relative to placebo, with associated 95% confidence limits. The size of each square reflects the weighting given to each response. The diamond at the base of each plot represents the overall effect calculated from a random effects model; the width of the diamond representing the 95% confidence interval.



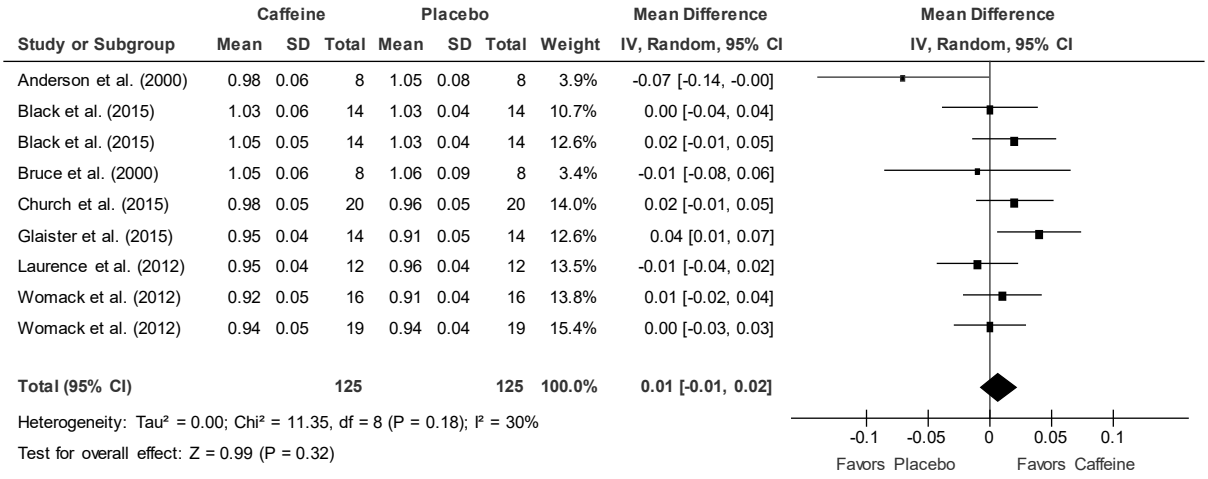


Figure 3. Forest plots of studies that have investigated the effects of caffeine supplementation on oxygen uptake (upper plot), and respiratory exchange ratio (lower plot) during closed-loop time-trial (≥ 5 mins) performance. Squares represent the raw mean difference, relative to placebo, with associated 95% confidence limits. The size of each square reflects the weighting given to each response. The diamond at the base of each plot represents the overall effect calculated from a random effects model; the width of the diamond representing the 95% confidence interval.

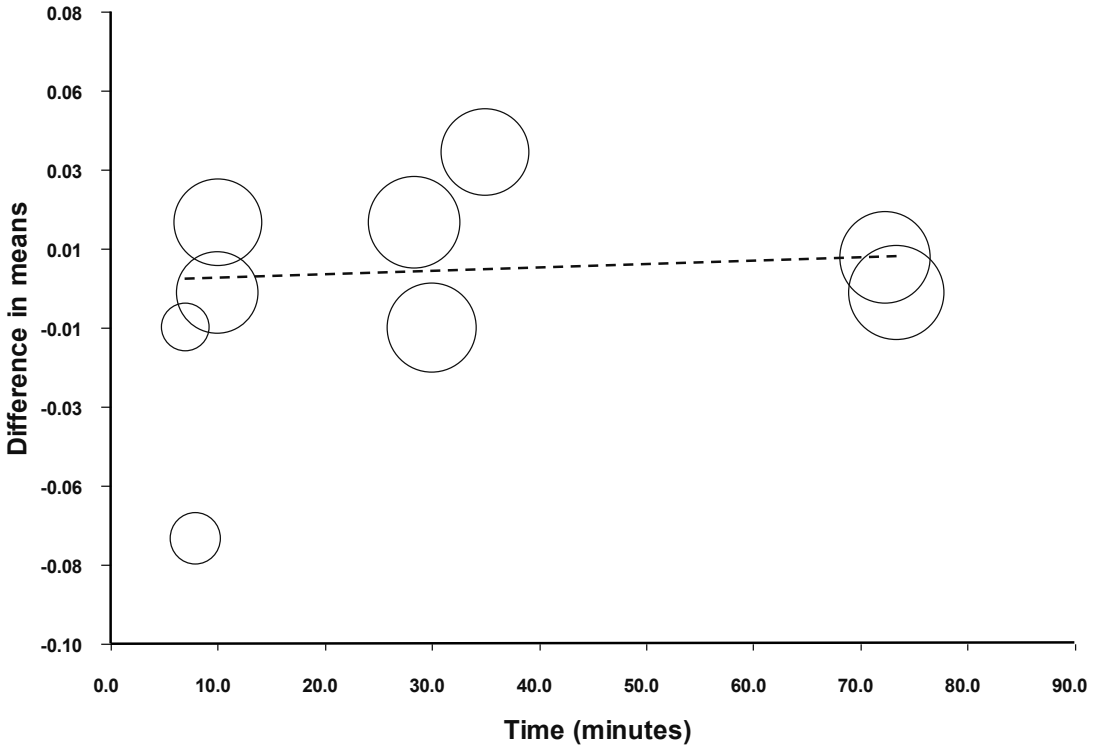


Figure 4. The relationship between exercise duration and the effect of caffeine on respiratory exchange ratio, relative to placebo, during closed-loop time-trial (≥ 5 mins) performance. Each circle represents an individual study, and the size of each circle is proportional to the weighting of each study in the analysis. The dashed line represents the line of best fit.

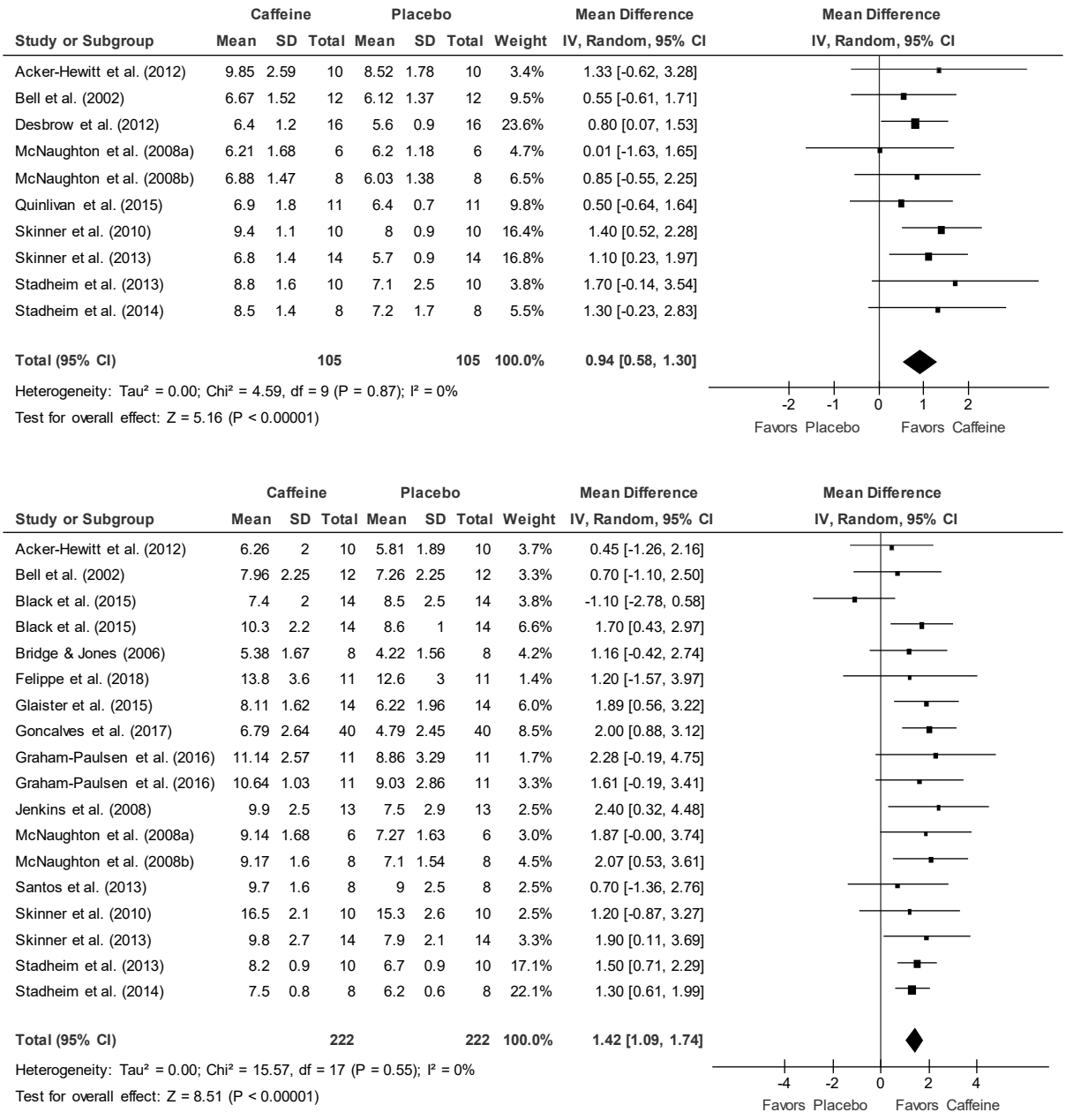


Figure 5. Forest plots of studies that have investigated the effects of caffeine supplementation on blood glucose (upper plot), and blood lactate (lower plot) concentrations during closed-loop time-trial (≥ 5 mins) performance. Squares represent the raw mean difference, relative to placebo, with associated 95% confidence limits. The size of each square reflects the weighting given to each response. The diamond at the base of each plot represents the overall effect calculated from a random effects model; the width of the diamond representing the 95% confidence interval.

Table 1. The effects of caffeine supplementation (3 – 6 mg·kg^{-1}), administered 30 – 90 minutes prior to exercise on closed-loop moderate to high-intensity (≥ 5 mins) time-trial performance and associated physiological responses.

Author(s)	n	Exercise mode	Time trial	Training status	Sex	Dose (mg·kg^{-1})	Pre-TT supplementation time (mins)	Supplementation method	Effect on TT	Physiological responses	PEDro score
Acker-Hewitt et al.[15]	10	Cycling	20 km	Cyclists	M	6	60	Capsule	no Δ in TT	no Δ in [BGl], [BLa], or RPE	8
Algrain et al.[27]	11[a]	Cycling	15 min	Recreational	M&F	~3.3	35	Gum	no Δ in TT	N/A	10
Algrain et al.[27]	9[b]	Cycling	15 min	Recreational	M&F	~3.3	35	Gum	no Δ in TT	N/A	10
Anderson et al.[21]	8	Rowing	2 km	Rowers	F	6	60	Capsule	no Δ in TT	no Δ in HR, RER, RPE, or $\dot{V}O_2$	10
Astorino et al.[29]	10	Cycling	8.2 km	Active	F	6	60	Drink[c]	↑ TT	no Δ in HR	10
Bell et al.[16]	12	Running	10 km	Runners	M&F	4	90	Capsule	no Δ in TT	↑ [BGl]; no Δ in [BLa] or HR	10
Berglund & Hemmingsson[30]	14	Skiing (Field)	~ 20 km	X-C skiers	M&F	6	60	Capsule	no Δ in TT	no Δ in RPE	8
Black et al.[31]	14	Cycling	10 min	Active	M&F	5	60	Capsule	↑ TT	↑ [BLa]; no Δ in HR, RER, RPE, or $\dot{V}O_2$	10
Black et al.[31]	14	Arm cranking	10 min	Active	M&F	5	60	Capsule	no Δ in TT	no Δ in HR, [BLa], RER, RPE, or $\dot{V}O_2$	10
Bortolotti et al.[32]	13	Cycling	20 km	Cyclists	M	6	60	Capsule	no Δ in TT	no Δ in HR or RPE	10
Boyett et al.[33]	20	Cycling	3 km	Various	M	6	60	Capsule	↑ TT	N/A	10
Bridge & Jones[34]	8	Running (Field)	8 km	Runners	M	3	60	Capsule	↑ TT	↑ [BLa] & HR; no Δ in RPE	10
Bruce et al.[22]	8	Rowing	2 km	Rowers	M	6	60	Capsule	↑ TT	↑ [BGl] & [BLa]; no Δ in HR, RER, RPE, or $\dot{V}O_2$	10
Christensen et al.[17]	12	Rowing	6 min	Rowers	M&F	3	45	Capsule	↑ TT	N/A	10
Church et al.[35]	20	Running	5 km	Recreational	M&F	3	60	Drink[e]	no Δ in TT	↑ RER; no Δ in [BGl], [BLa], HR, or $\dot{V}O_2$	10
Desbrow et al.[23]	16	Cycling	~ 60 min	Cyclists	M	6	90	Capsule	↑ TT	↑ [BGl] & HR; no Δ in RPE	10
Felippe et al.[36]	11	Cycling	4 km	Cyclists	M	5	75	Capsule	↑ TT	no Δ in [BLa], HR, RPE, or $\dot{V}O_2$	10
Giersch et al.[37]	20	Cycling	3 km	Cyclists	M	6	60	Capsule	no Δ in TT	N/A	9
Glaister et al.[18]	14	Cycling	20 km	Cyclists/Triathletes	F	5	60	Capsule	↑ TT	↑ [BLa], HR, & RER; no Δ in RPE or $\dot{V}O_2$	10
Gonçalves et al.[38]	40	Cycling	~ 30 min	Cyclists	M	6	60	Capsule	↑ TT	no Δ in [BLa] or RPE	10
Graham-Paulsen et al.[39]	11	Cycling	10 km	Recreational	M	4	90	Capsule	↑ TT	↑ [BLa]; no Δ in RPE	9
Graham-Paulsen et al.[39]	11	Hand cycling	10 km	Recreational	M	4	90	Capsule	no Δ in TT	↑ [BLa]; no Δ in RPE	9
Hodgson et al.[40]	8	Cycling	45 min	Cyclists/Triathletes	M	5	60	Drink[d]	↑ TT	no Δ in HR	8
Hodgson et al.[40]	8	Cycling	45 min	Cyclists/Triathletes	M	5	60	Drink[e]	↑ TT	no Δ in HR	8
Irwin et al.[41]	12	Cycling	~ 60 min	Cyclists	M	3	90	Capsule	↑ TT	↑ HR; no Δ in RPE	10
Jenkins et al.[24]	13	Cycling	15 min	Cyclists	M	3	60	Capsule	no Δ in TT	↑ [BLa]	8
Lane et al.[19]	12	Cycling	44 km	Cyclists/Triathletes	M	3	40	Gum	↑ TT	no Δ in HR	10
Lane et al.[19]	12	Cycling	29 km	Cyclists/Triathletes	F	3	40	Gum	↑ TT	no Δ in HR	10
Laurence et al.[42]	12	Cycling	30 min	Sedentary	M	6	60	Capsule	↑ TT	↑ HR & $\dot{V}O_2$; no Δ in RER or RPE	10
McNaughton et al.[43]	6	Cycling	60 min	Cyclists	M	6	60	Drink[c]	↑ TT	no Δ in [BGl], [BLa], or HR	10
McNaughton et al.[44]	8	Cycling	60 min	Cyclists	M	6	60	Drink[c]	↑ TT	no Δ in [BGl], [BLa], or HR; ↓ RER	10
Quinlivan et al.[20]	11	Cycling	~ 60 min	Cyclists	M	3	90	Capsule	↑ TT	no Δ in [BGl], HR, or RPE	10
Santos et al.[45]	8	Cycling	4 km	Cyclists	M	5	60	Capsule	↑ TT	no Δ in [BLa], HR, RPE, or $\dot{V}O_2$	10
Saunders et al.[46]	42	Cycling	~30 min	Cyclists	M	6	60	Capsule	↑ TT	N/A	10
Skinner et al.[25]	10	Rowing	2 km	Rowers	M	6	60	Capsule	no Δ in TT	↑ [BGl] & [BLa]; no Δ in RPE or $\dot{V}O_2$	10
Skinner et al.[47]	14	Cycling	40 km	Cyclists/Triathletes	M	6	60	Capsule	↑ TT	↑ [BGl]; no Δ in [La] or RPE	10
Stadheim et al.[48]	10	X-C skiing	8 km	X-C skiers	M	6	75	Drink[c]	↑ TT	↑ [BGl], [BLa], & HR	10
Stadheim et al.[49]	8	X-C skiing	10 min	X-C skiers	M	4.5	75	Drink[c]	↑ TT	↑ [BGl], [BLa], & HR	10
Wallman et al.[50]	10	Cycling	10 min	Sedentary	F	6	60	Capsule	no Δ in TT	no Δ in HR, RER, RPE, or $\dot{V}O_2$	10
Womack et al.[51]	16[a]	Cycling	40 km	Cyclists	M	6	60	Capsule	↑ TT	↑ HR & $\dot{V}O_2$; no Δ in RER	10
Womack et al.[51]	19[b]	Cycling	40 km	Cyclists	M	6	60	Capsule	↑ TT	↑ HR & $\dot{V}O_2$; no Δ in RER	10

Note: ↑, significant ($p < 0.05$) increase relative to placebo; ↓, significant ($p < 0.05$) decrease relative to placebo; [BGl], end-test blood glucose concentration; [BLa], end-test blood lactate concentration; F, female; HR, mean heart rate; M, male; no Δ, no significant ($p \geq 0.05$) change relative to placebo; PEDro, Physiotherapy evidence database scale; RER, mean respiratory exchange ratio; RPE, end-test rating of perceived exertion; TT, time-trial (↑ TT, improved time-trial performance relative to placebo); $\dot{V}O_2$, mean rate of oxygen consumption; X-C, cross country; [a], AA homozygotes (CYP1A2 gene); [b], C allele carriers (CYP1A2 gene); [c], dose added to artificially sweetened water/lemonade/juice. [d], dose added to water; [e], dose served as coffee.

Table 2. Summary of subgroup meta-analyses examining the possible influence of supplementation method (capsule vs drink formats) and caffeine dose (≥ 5 mg·kg^{-1} vs < 5 mg·kg^{-1}) on the effect of caffeine supplementation on mean respiratory exchange ratio during moderate- to high-intensity closed-loop time-trial performance.

Responses	**No of studies**	**Sample size**	**Mean difference**	***P***	**Heterogeneity I^2 (%)**	**Subgroup differences**	
						I^2 (%)	***p***
Respiratory exchange ratio							
Capsule	8	105	.00 [-.01, .02]	.55	35 [0, 71]	0	.40
Drink	1	20	.02 [-.01, .05]	.21	N/A*		
≥ 5 mg·kg^{-1}	8	105	.00 [-.01, .02]	.55	35 [0, 71]	0	.40
< 5 mg·kg^{-1}	1	20	.02 [-.01, .05]	.21	N/A*		

Note: Values in square brackets represent 95% confidence limits; *unable to be calculated due to an insufficient number of studies.