Fasting before evening exercise reduces net energy intake and increases fat oxidation, but impairs performance in healthy males and females

Authors: Tommy Slater[1], William J A Mode[1], Mollie G Pinkney[1], John Hough[1], Ruth M James[1], Craig Sale[1,2], Lewis J James[3] and David J Clayton[1*]

Affiliations: [1]*Musculoskeletal Physiology Research Group, Sport, Health and Performance Enhancement Research Centre, School of Science and Technology, Nottingham Trent University, Nottingham, UK.*

[2]*Institute of Sport, Manchester Metropolitan University, Manchester, M1 7EL, UK*

[3]*National Centre for Sport and Exercise Medicine, School of Sport, Exercise and Health Sciences, Loughborough University, Loughborough, Leicestershire, LE11 3TU, UK.*

Running Head: Fasting, evening exercise and energy intake

***Corresponding author:** Dr David J Clayton

Senior Lecturer in Nutrition and Exercise Physiology

Sport, Health and Performance Enhancement Research Centre, School of Science and Technology; Clifton Campus; Nottingham Trent University; Nottingham; Nottinghamshire; NG11 8NS; UK

Email: David.Clayton@ntu.ac.uk

Tel: +44 (0) 115 84 85514

Word count: 4222

ABSTRACT

Acute morning fasted exercise may create a greater negative 24-h energy balance than the same exercise performed after a meal, but research exploring fasted evening exercise is limited. This study assessed the effects of 7 h fasting before evening exercise on energy intake, metabolism, and performance. Sixteen healthy males and females (*n*=8 each) completed two randomised, counterbalanced trials. Participants consumed a standardised breakfast (08:30) and lunch (11:30). Two hours before exercise (16:30), participants consumed a meal (543 ± 86 kcal; FED) or remained fasted (FAST). Exercise involved 30 min cycling (60% $\dot{V}O_{2peak}$) and a 15-min performance test (~85% $\dot{V}O_{2peak}$; 18:30). *Ad-libitum* energy intake was assessed 15 min post-exercise. Subjective appetite was measured throughout. Energy intake was 99 ± 162 kcal greater post-exercise ($P<0.05$), but 443 ± 128 kcal lower over the day ($P<0.001$), in FAST. Appetite was elevated between the pre-exercise meal and *ad-libitum* meal in FAST ($P<0.001$), with no further differences ($P\geq0.458$). Fat oxidation was greater (+3.25 ± 1.99 g), and carbohydrate oxidation lower (-9.16 ± 5.80 g) during exercise in FAST ($P<0.001$). Exercise performance was 3.8% lower in FAST (153 ± 57 vs. 159 ± 58 kJ; $P<0.05$), with pre-exercise motivation, energy, readiness, and post-exercise enjoyment also lower in FAST ($P<0.01$). Fasted evening exercise reduced net energy intake and increased fat oxidation compared to exercise performed 2 h after a meal. However, fasting also reduced voluntary performance, motivation, and exercise enjoyment. Future studies are needed to examine the long-term effects of this intervention as a weight management strategy.

Keywords: Exercise performance, appetite, energy intake, energy balance, fasting, fat oxidation

INTRODUCTION

Weight gain occurs when energy intake exceeds energy expenditure for a sustained period (Hill et al., 2012). Counter-regulatory changes to energy balance systems appear more profound for weight loss than weight gain (Hill et al., 2012), meaning early intervention in lean individuals to prevent weight gain might be a more efficacious approach than attempting to reduce obesity once established (Monnier et al., 2021).

Physical activity/exercise can aid weight management by increasing energy expenditure (Donnelly et al., 2009). Despite this, long-term exercise interventions for weight management are often less effective than predicted (Martin et al., 2019), perhaps explained by compensatory reductions in energy expenditure (Thompson et al., 2014), and/or increases in energy intake (King et al., 2008). Manipulating the timing of exercise around meals has the potential to optimise exercise as a strategy to sustain an energy deficit and/or improve metabolic health.

Exercise after a prolonged fast (>12 h) may aid in regulating energy balance. For example, compared to consuming breakfast before exercise, fasted exercise produces either no change (Bachman et al., 2016; Gonzalez et al., 2013; Griffiths et al., 2020) or a small increase (Edinburgh et al., 2019) in lunch energy intake, but facilitates a lower overall energy intake (breakfast plus lunch), and lowers 24 h energy intake (Bachman et al., 2016; Edinburgh et al., 2019). Additionally, fasted morning exercise increases fat oxidation (Edinburgh et al., 2019; Gonzelez et al., 2013), which may drive adaptations leading to improved markers of metabolic health (Robinson et al., 2015).

Almost all research on fasted exercise has been undertaken in the morning because the overnight fast offers a practical and convenient opportunity to achieve a fasted state without the need to skip meals. The response to fasted exercise at other times of day is not well researched. There is evidence that evening exercise is associated with a reduced perception of effort (Maraki et al., 2005), and may improve glycaemic control more than morning exercise (Moholdt et al., 2021). These diurnal differences may be explained by the circadian system, which regulates several endogenous processes, including macronutrient metabolism, appetite, and components of energy balance, in 24 h oscillations (Smith and Betts, 2022). Therefore, findings from overnight-fasted exercise might not translate to exercise performed later in the day.

Only one study has examined the energy intake responses to fasted-state exercise performed at a time of day other than the morning. McIver et al. (2019a) showed similar 24 h energy intakes following fed- and fasted-state exercise commencing in the morning or early-evening, indicating fasted exercise may reduce daily energy intake, irrespective of the time of day. The amount of exercise performed and motivation to exercise are, however, important to maximise both the energy deficit achieved and the positive health outcomes from exercise training (Foulds et al., 2014). Skipping breakfast has been shown to reduce voluntary exercise performance (Clayton and James, 2016), but the effect of fasting on evening exercise performance is unknown.

The present study examined the effects of fasting for 7 h before evening cycling exercise on post-exercise *ad-libitum* energy intake, appetite, voluntary exercise performance, and substrate oxidation in healthy, recreationally active males and females.

METHODS

Participants

Sixteen healthy, recreationally active (<10 h·wk^{-1}) males and females (n=8 each) completed the study (Table 1), which was conducted in accordance with the Declaration of Helsinki and approved by the Nottingham Trent University Ethical Advisory Committee; ethics application number: 670. Clinical Trials registration: NCT04742530. Herein, we describe the first of two studies completed as part of this clinical trial. A separate study will be published elsewhere comparing the effects of fed-state evening exercise (including the data from 15 participants presented here) to fed-state morning exercise. Participants were not restrained, disinhibited, or hungry eaters, determined by the three-factor eating questionnaire (Stunkard and Messick, 1985). Female participants were regular monophasic combined oral contraceptive users (≥6 months use before commencing the study; *n*=3) or eumenorrheic (self-reported; *n*=5), and not using a hormonal contraceptive. Participants completed health screening questionnaires and provided written informed consent before commencing the study. The sample size for this study was estimated for the primary outcome variables of voluntary exercise performance and energy intake using G*Power 3.1 software. Using an α of 0.05, β of 0.8 and data from a previous study (Clayton et al., 2015), it was estimated that 15 participants would be required to detect a 5% difference in voluntary exercise performance, and 12 participants to detect a 15% difference in energy intake. Secondary outcome variables include substrate oxidation, appetite, and subjective responses to exercise.

Study design

Participants completed two preliminary trials, followed by two experimental trials (completed between February–July 2021 in Nottingham Trent University laboratories) in randomised (by drawing trial orders for participants out of a bag), counterbalanced, cross-over order, and were separated by ≥4 days. To control for fluctuations in appetite associated with sex hormone concentrations (Buffenstein et al., 1995), eumenorrheic women completed experimental trials in the follicular phase (3–14 days after the onset of menstruation – self-reported) and oral contraceptive users completed all trials between days 4–17 of the pill-taking phase. This was individually standardised within a 4-day period for each female participant. Experimental trials involved consuming a 24 h standardised diet before an exercise session at 18:30. Exercise consisted of 30-min steady-state cycling and a 15-min all-out performance test, which required participants to complete as much work as possible within the allotted time. In FAST, participants ceased food intake at 11:30 and commenced exercise after a 7 h fast. In FED, participants consumed a pre-exercise meal at 16:30 and commenced exercise after a 2 h fast.

Preliminary trials

During the first preliminary trial, participants' body mass and height were measured, before body fat percentage was estimated by measuring skinfold thickness (Durnin and Womersley, 1974). Cycling $VO_{2\text{peak}}$ was determined during a discontinuous incremental exercise test on an electronically braked cycle ergometer (Lode Corival, Netherlands). The test involved 4-min incremental stages separated by ~5 min rest until volitional exhaustion. Heart rate, rating of perceived exertion (RPE; Borg, 1982), and 1-min expired gas samples were collected during the final minute of each increment. After adequate rest, participants completed the 15-min performance test. During the second preliminary trial, participants were familiarised with the exercise protocol and the *ad-libitum* meal.

Pretrial standardisation

Participants recorded food intake and habitual physical activity during the 24 h prior to the first experimental trial and replicated this before the second experimental trial. Strenuous physical activity and alcohol intake were prohibited during this period, with adherence confirmed verbally before each trial. Participants arrived at the laboratory via motorised transport.

Protocol

Participants consumed a standardised dinner at 20:30 the evening before trial days, a breakfast at 08:30, and a lunch at 11:30. In FED, participants consumed a standardised pre-exercise meal at 16:30, which was replaced with a prescribed volume of water in FAST. Participants arrived at the laboratory at 18:00 and measures of subjective appetite, mood, and exercise readiness were completed. After 20 min supine rest, a 5-min expired gas sample was collected. Exercise commenced at 18:30, with 30-min steady-state cycling (60% VO_{2peak}). During exercise, heart rate and RPE were measured every 5 min, with 2-min expired gas samples collected every 10 min. After 3-min rest, participants commenced a 15-min all-out performance test. An *ad-libitum* pasta meal was served 15 min after the cessation of exercise, and participants were permitted 20 min to eat. Participants then left the laboratory and were instructed to consume nothing other than the prescribed water and to refrain from engaging in exercise until after completing the final subjective appetite questionnaire at 08:30 the following day. Adherence to this was confirmed via text messaging.

Exercise performance test

The ergometer was set in linear mode, with the linear factor (L) calculated using the formula: $L = W/(\text{rpm})^2$ to elicit a workload (W) of 85% VO_{2peak} at the participants' preferred cadence identified during the VO_{2peak} test. Power output could be increased and decreased with an increase or decrease in cadence. Participants completed as much work as possible within 15 min and were blinded to all outcome measures, except time remaining. No encouragement was provided, and standardised instructions were provided before each trial. Work completed (kJ) and heart rate were recorded every minute, and RPE was recorded every 2 min from the first minute.

Standardised meals

Participants were provided with weighed meals and water to be consumed at home, with clear, written guidelines on timing of intake and instruction to consume nothing else. Participants were regularly contacted via text messaging to encourage adherence with these instructions. Meals were designed to provide a percentage of estimated energy requirements (EER; resting metabolic rate [Mifflin et al., 1990] multiplied by a physical activity level of 1.7).

Standardised dinner and lunch meals were identical (30% EER), consisting of tuna/chicken sandwiches prepared by the researchers (white bread (Hovis, UK), tuna chunks in brine (Princess, UK)/chicken breast chunks (Bernard Matthews, UK), and full-fat mayonnaise (Hellmann's, UK)), ready salted crisps (Walkers, UK), and chocolate (Cadbury, UK). Standardised breakfast and pre-exercise meals were also identical (20% EER), consisting of instant porridge oats (Oatso Simple Golden Syrup, Quaker, UK), cereal bars (Strawberry Nutri-Grain, Kellogg's, UK), and yoghurt (Ski Strawberry, Nestlé, UK) (Table 2). Water intake was provided at 30 $\mathrm{mL \cdot kg^{-1}}$ body mass during trials, distributed into 5 equal volumes consumed: 1) between waking and lunch (<11:30); 2) during lunch (11:30–12:00); 3) early afternoon (12:00–17:30); 4) 1 h before exercise (17:30); and 5) between the *ad-libitum* meal and sleep (>20:00).

***Ad-libitum* meal**

Energy and water intake were determined by weighing food and water before and after consumption. The *ad-libitum* meal was homogenous, providing 1.25 ± 0.01 $\mathrm{kcal \cdot g^{-1}}$ (69% carbohydrate, 11% protein, 18% fat, and 2% fibre), and consisted of pasta, tomato sauce and olive oil. The meal was provided in excess of expected consumption, and participants ate in isolation to eliminate distractions until they felt *"comfortably full and satisfied"*. Water was available *ad-libitum*. Participants remained in the booth for the 20-min period, and all participants reported they had ceased eating within this time in all trials.

Expired gas samples

A 5-min expired gas sample was collected into a Douglas bag immediately pre-exercise following 20 min of supine rest. During steady-state cycling, 2-min expired gas samples were collected between 8–10, 18–20, and 28–30 min. Samples were assessed for oxygen and carbon dioxide concentrations (MiniHF 5200, Servomex, UK), volume (Harvard Dry Gas Meter, Harvard Ltd., UK), and temperature. Substrate oxidation rates were calculated using stoichiometric equations (Jeukendrup and Wallis, 2005).

Subjective responses

Participants rated their subjective feelings of hunger, fullness, desire to eat (DTE), prospective food consumption (PFC), and nausea on digital visual analogue scales (VAS) that were sent to their personal mobile telephone at each timepoint (0, 2, 3, 3.5, 5, 7, 8, 10, 11, 11.5, 13.5, and 24 h). Additional subjective feelings of motivation to exercise, readiness to exercise, tiredness, and energy were added to the pre-exercise questionnaire (10 h). All VAS were designed and administered using SurveyMonkey.com and comprised of a 0 to 100 sliding scale with written anchors of “not at all”/“no desire at all”/“none at all” and “extremely”/“a lot” placed at 0 and 100, respectively. Participants also completed a paper-based Positive and Negative Affect Schedule (PANAS; Watson et al., 1988) pre-exercise.

A paper-based, shortened version of the Physical Activity Enjoyment Scale (PACES-8) was completed immediately post-exercise to measure enjoyment of exercise sessions (Raedeke, 2007). The PACES-8 uses a series of eight, seven-point bipolar scales which participants use to rate their agreement with one of the two statements at either end of the scale (*e.g.,* “I enjoyed it” – “I hated it”).

Statistical analyses

Data were analysed using SPSS v26.0 (IBM, USA). All data were checked for normality of distribution using a Shapiro-Wilk test. For subjective appetite-related variables, area under the curve (AUC) values were calculated using the trapezoidal method and averaged over time in response to breakfast (0–3 h), lunch (3–7 h), pre-exercise meal (7–11 h), and *ad-libitum* meal (11–24 h). Data containing one factor were analysed using paired samples *t*-tests or Wilcoxon Signed-Rank tests as appropriate. Data containing two factors were analysed using repeated-measures ANOVA, with significant main effects followed by *post-hoc* paired samples *t*-tests, or Wilcoxon Signed-Rank tests, with Holm-Bonferroni correction. Because fluctuations in circulating sex hormone concentrations can influence appetite and energy intake in females, sex was entered as a between-participants factor in repeated-measures ANOVA to test for sex-by-trial-by-time interactions and/or sex-by-trial interactions. Due to equipment issues, heart rate data is missing for one participant. Data sets were considered statistically different when $P < 0.05$. Data are presented as mean ± 1SD, unless stated. Where appropriate, effect sizes (Cohen’s *dz*) were calculated (Cohen, 1988).

RESULTS

Energy intake

Ad-libitum energy intake post-exercise was 99 ± 162 kcal greater during FAST ($dz = 0.61$; $P < 0.05$), but cumulative energy intake across the day was 443 ± 128 kcal lower during FAST than FED ($dz = 3.42$; $P < 0.001$; Table 2).

There was a sex-by-trial interaction effect for *ad-libitum* energy intake ($P < 0.001$), with greater energy intake during FAST than FED in males (+203 ± 122 kcal; $dz = 1.67$; $P < 0.01$), but not females (-5 ± 129 kcal; $dz = 0.04$; $P = 0.919$; Figure 1).

Subjective appetite responses

There were trial ($P < 0.01$) and time ($P < 0.01$) main effects and a trial-by-time interaction ($P < 0.001$) effect for hunger, fullness, DTE, and PFC. Participants reported increased hunger, DTE, and PFC, and reduced fullness, in the period following the pre-exercise meal until immediately before the post-exercise *ad-libitum* meal (16:30–19:30) during FAST ($P < 0.05$). Nausea showed a main effect of time ($P < 0.01$), and a trial-by-time interaction effect ($P < 0.05$), but no main effect of trial ($P = 0.149$). Nausea tended to be greater immediately pre-exercise in FAST ($P = 0.06$; Figure 2).

AUC for hunger, DTE, PFC, and nausea were all greater, and fullness was lower, between the pre-exercise meal and the *ad-libitum* meal in FAST ($P < 0.01$). No further AUC differences were shown between trials in response to breakfast ($P \geq 0.398$), lunch ($P \geq 0.458$) or *ad-libitum* meal ($P \geq 0.464$; Figure 3).

Energy expenditure and substrate oxidation

At rest, carbohydrate oxidation was lower (0.04 ± 0.03 vs 0.13 ± 0.06 $g \cdot min^{-1}$; $dz = 1.25$; $P < 0.001$) and fat oxidation was higher (0.11 ± 0.02 vs 0.09 ± 0.03 $g \cdot min^{-1}$; $dz = 0.67$; $P < 0.01$) in FAST. Energy expenditure at rest was lower in FAST (1.3 ± 0.2 $kcal \cdot min^{-1}$ vs 1.2 ± 0.2 $kcal \cdot min^{-1}$; $dz = 0.67$; $P < 0.001$). There was a sex-by-trial interaction effect for resting energy expenditure ($P < 0.05$), which was lower in FAST than FED in males (1.5 ± 0.2 $kcal \cdot min^{-1}$ vs 1.3 ± 0.2 $kcal \cdot min^{-1}$; $dz = 1.12$; $P < 0.05$) but was not different between trials in females (1.2 ± 0.1 $kcal \cdot min^{-1}$ vs 1.1 ± 0.1 $kcal \cdot min^{-1}$; $dz = 0.14$; $P = 0.602$).

During steady-state exercise, total fat oxidation was greater (+3.25 ± 1.99 g; *dz* = 1.64; $P < 0.001$), and total carbohydrate oxidation was lower (-9.16 ± 5.80 g; *dz* = 1.58; $P < 0.001$) in FAST (Figure 4). Total energy expenditure in the steady-state exercise was lower in FAST (-6 ± 8 kcal; *dz* = 0.59; $P < 0.05$).

Exercise performance and responses

Work completed during the 15-min performance test was 5 ± 8 kJ lower during FAST (*dz* = 0.62; $P < 0.05$; Figure 5).

Mean VO_2 achieved during steady-state exercise was lower in FAST (57.9 ± 5.6% VO_{2peak} vs. 59.0 ± 6.1% VO_{2peak}; $P < 0.01$). Mean heart rate ($P = 0.079$) and RPE ($P = 0.806$) were not different between trials during the 30-min steady-state bout. Mean heart rate during the performance test was lower in FAST ($P < 0.05$), but RPE was not different between trials ($P = 0.739$).

Laboratory temperature ($P = 0.212$), humidity ($P = 0.702$), and pressure ($P = 0.442$) were not different between trials.

Exercise subjective responses

Participants reported lower pre-exercise motivation, energy, and readiness to exercise in FAST ($P < 0.001$), although tiredness was not different between trials ($P = 0.270$). The PANAS questionnaire revealed lower positive affect pre-exercise in FAST ($P < 0.05$), but negative affect was not different between trials ($P = 0.238$). Mean score on the PACES-8 questionnaire was lower in FAST ($P < 0.01$), suggesting that the exercise session was enjoyed less in FAST (Table 3).

DISCUSSION

We showed that fasting for 7 h before evening exercise increased *ad-libitum* energy intake by ~100 kcal compared to exercise performed 2 h after eating, but this did not compensate for the omission of a pre-exercise meal. Accordingly, net energy intake was lower when evening exercise was performed following a 7 h fast. However, fasting before evening exercise reduced performance by ~3.8%, and was associated with reduced motivation and exercise enjoyment. Further study is required to determine whether fasting before evening exercise can be used chronically to assist in weight and health management, or whether it's associated negative perceptions impede long-term success.

Most studies explore fasted-state exercise in the morning due to the convenience of extending the overnight fast. However, morning exercise is not always convenient or possible, so this study assessed the metabolic and behavioural responses to fasted-state exercise in the evening. Previously, McIver et al. (2019a) showed that fasting for 9 h before exercising at 17:00 increased appetite pre-exercise, but post-exercise appetite was not different to fed-state exercise. This aligns with some (Gonzalez et al., 2013; McIver et al., 2019b), but not all (Bachman et al., 2016; Griffiths et al., 2020) morning fasted-state exercise studies. Findings from the present study are in-line with the latter, demonstrating elevated appetite extending into the post-exercise period. Interestingly, post-exercise energy intake was ~100 kcal (~13%) greater, which contrasts the results of studies where exercise is performed in the morning (Bachman et al., 2019; Gonzalez et al., 2013; Griffiths et al., 2020). As such, the present study provides novel data suggesting a potential disparity in post-exercise energy intake responses between morning and evening fasted-state exercise, with evening fasted-state exercise appearing to provoke compensatory eating which is not typically found with morning fasted-state exercise, although further studies directly comparing morning and evening fasted-state exercise are still needed.

Interestingly, this increase in energy intake was driven predominantly by males, with seemingly no such compensation occurring in females. Appetite and energy intake responses to acute exercise are generally similar between males and females (Dorling et al., 2018), although only a small number of studies have directly compared males and females. Moreover, nutrient-exercise interactions have not been considered (Frampton et al., 2022), so the sex-specific responses to fasted-state exercise are unknown. Our findings suggest that fasted-state evening exercise may provoke a smaller compensatory energy intake response in females, potentially making it a more effective weight management strategy for females than males. Sex hormones may influence appetite and energy intake (Buffenstein et al., 1995). We attempted to control this by conducting trials in the same phase of the menstrual or pill-taking cycle. However, we were unable to standardised this to the exact day within the phase, and we did not measure hormones directly, both of which can be considered limitations of the present study. Sex hormone concentrations may still fluctuate within the same cycle phase (Buffenstein et al., 1995), meaning larger sample size studies of both males and females with measurement of ovarian hormone concentrations are required to further explore these preliminary findings.

Despite post-exercise energy intake being greater following fasted-state evening exercise, this increase only compensated for ~18% of the pre-exercise meal in FED. Therefore, energy intake over the course of the entire day was ~443 kcal lower in the fasted trial. Energy intake was only measured at a single post-exercise meal, so it is possible that further energy intake compensation may occur later in the evening or during the subsequent day. Consistent with other studies (Bachman et al., 2016; McIver et al., 2019a; McIver et al., 2019b; Griffiths et al., 2020), differences in appetite were abolished after the post-exercise meal, implying that future eating behaviour may not differ between trials. Indeed, studies tracking energy intake for up to 24 h post-exercise demonstrate that the reduction in energy intake caused by fasting (meal skipping) is not compensated for in this time period (McIver et al., 2019a; Edinburgh et al., 2019; Bachman et al., 2016). Additionally, recent work suggests energy intake increases in anticipation of energy restriction (James et al., 2020) and/or exercise (Barutcu et al., 2021), but this could not be assessed in the present study, as food intake was controlled to ensure similar metabolic conditions at the start of trials.

We showed that prior fasting for 7 h increased fat oxidation by 3.25 g during 30 min evening exercise. Exercising after a 10–14 h overnight fast increases fat oxidation (Edinburgh et al., 2019; Gonzalez et al., 2013), which if performed regularly, may drive adaptations leading to improved markers of metabolic health (Robinson et al., 2015). Despite circadian variations in several metabolic processes (Smith and Betts, 2022), the present study, and previous work (McIver et al., 2019a), show that a shorter, 7–9 h fasting period during the afternoon also increases fat oxidation during evening exercise. However, it must be noted that longer fasting durations that include the overnight fast, and shorter fasting durations such as that used in the present study, likely elicit differences in metabolism beyond changes in substrate oxidation. For example, plasma glycerol concentrations (a marker of lipolysis) increase in direct proportion to the duration of the fast (Montain et al., 1991), meaning the metabolic effects of a shorter period of afternoon fasting may not necessarily mimic those of an overnight fast. Future studies should seek to explore whether elevated fat oxidation during fasted-state evening exercise improves markers of metabolic health.

The main benefits from exercise are likely to be driven by the volume and intensity of exercise performed (Foulds et al., 2014). This is especially important when time for exercise is often curtailed by other commitments (Cerin et al., 2010). We found that fasting before evening exercise reduced subjective ratings of motivation, readiness, and energy immediately prior to exercise, indicating a suboptimal psychological state for maximising the volume or intensity of voluntary exercise. Accordingly, the amount of work completed during the 15-

min performance test was reduced by 3.8% with fasting. Eating, particularly carbohydrate, appears to enhance aerobic performance >60 min due partially to increased endogenous carbohydrate stores (Aird et al., 2018), but effects on aerobic exercise <60 min are less conclusive (Mears et al., 2018; Galloway et al., 2014). Recent evidence suggests that the perception of consuming nutrients prior to exercise using an energy-free "placebo" meal (Mears et al., 2018; Naharudin et al., 2020) or the suppression of hunger (Naharudin et al., 2021), might improve performance. Therefore, the awareness of consuming nutrients and/or subjective responses during the fed-state exercise trial may have increased self-selected intensity during the performance test.

The absolute difference between trials for work completed was very small (~6 kcal), possibly due to the short duration (15 min) and high intensity (85% VO_{2peak}) of the selected test. This reduction in performance is unlikely to manifest in meaningful change to energy balance. However, if motivation to exercise and self-selected duration and/or intensity of exercise are curtailed, as this reduction in performance might imply, this could dramatically impact the success of exercise training programmes. Additionally, given that exercise enjoyment may be an important predictor of long-term adherence to exercise interventions (Raedeke, 2007), the finding of reduced exercise enjoyment in the present study provides further insight into possible challenges with incorporating fasted-state evening exercise into a weight management programme.

The present study provides novel insight into the effects of fasting before evening exercise, but it is not without limitations. Firstly, the absence of an overnight-fasted trial precludes the direct comparison of morning and evening fasted-state exercise. Secondly, participants were required to consume standardised meals and undergo instructed fasting periods in the absence of experimenter supervision. Although regular contact was made via text messaging to increase compliance, full adherence with these instructions cannot be assumed. Thirdly, the study was conducted in lean and healthy participants, meaning the results cannot be directly extrapolated to other population groups, particularly individuals with overweight or obesity, who may respond differently to fasting-based interventions (Gonzalez et al., 2018). Finally, this study investigated a single exposure, and compensatory energy intake was only assessed at a single timepoint. As such, it is not known whether our acute findings would persist after multiple exposures within a free-living setting, with greater opportunity for compensatory energy balance behaviours to occur.

CONCLUSION

This study showed that fasting for 7 h prior to evening exercise may be an effective method of reducing net energy intake, whilst also increasing fat oxidation. The chronic success of this intervention may, however, be compromised by elevations in appetite and reductions in voluntary performance, as well as reductions in the motivation to exercise and the enjoyment of exercise sessions. Future studies are required to explore whether regular fasted-state evening exercise can be used by lean and healthy individuals as a method of managing body weight and/or composition in the long-term. Additionally, exploring the effects of this intervention on indices of energy balance and metabolic health within overweight/obese populations represents an important avenue for future research.

Acknowledgments

The authors would like to thank Terrance Campion and Beverley Armstrong for their assistance during data collection.

Authorship

All authors contributed to the study conception and design; Tommy Slater, William J A Mode, Mollie G Pinkney and David J Clayton completed data collection; Tommy Slater, William J A Mode and David J Clayton analysed the data; the first draft of the manuscript was written by Tommy Slater; and all authors critically reviewed previous versions of the manuscript. All authors have read and approved the final manuscript.

Conflict of interest

LJJ is part of the National Institute for Health Research's Leicester Biomedical Research Centre, which is a partnership between University Hospitals of Leicester NHS Trust, Loughborough University, and the University of Leicester. This report is independent research by the National Institute for Health Research. The views expressed in this publication are those of the authors and not necessarily those of the NHS, the National Institute for Health Research, or the Department of Health. LJJ has current/previous funding from Entrinsic Beverage Company LLP, Herbalife Europe Ltd, Bridge Farm Nurseries, Decathlon SA, PepsiCo Inc., Volac International, has performed consultancy for PepsiCo Inc. and Lucozade, Ribena Suntory, and has received conference fees from PepsiCo Inc. and Danone Nutricia. In all cases, monies have been paid to LJJs institution and not directly to LJJ.

CS has no conflicts of interest to declare as they relate directly to the topic of this study. More generally, potential and perceived conflicts of interest over the last few years include: Research funding from the UK Ministry of Defence, Natural Alternatives International, English Institute of Sport, NHS Nottingham City, Birmingham City University, Coventry University and GlaxoSmithKline HPL (all as PI) and Fundação de Amparo à Pesquisa do Estado de São Paulo (Brazil), Ciência sem Fronteiras (Brazil), British Milers Club, Irish Research Council and NHS Nottingham City (as Co-I). Honoraria have been received from the Gatorade Sport Science Institute, UK Dairy Council, Guru Performance Ltd, International Society of Sports Nutrition, English Institute of Sport, GlaxoSmithKline HPL and Nutrition X. Other 'in kind' research support has been received from Natural Alternatives International

in the form of supplements for research, support to attend a conference and payment of open access page charges.

Funding Sources

Tommy Slater is supported by a PhD studentship awarded by Nottingham Trent University.

Protocol

Trial registration: 8th February 2021 (https://clinicaltrials.gov/ct2/show/NCT04742530)

References

Aird, T. P., Davies, R. W., & Carson, B. P. (2018). Effects of fasted vs fed-state exercise on performance and post-exercise metabolism: A systematic review and meta-analysis. Scandinavian Journal of Medicine & Science in Sports, 28(5), 1476–1493. https://doi.org/10.1111/sms.13054

Bachman, J. L., Deitrick, R. W., & Hillman, A. R. (2016). Exercising in the fasted state reduced 24-hour energy intake in active male adults. Journal of Nutrition and Metabolism 2016. https://doi.org/10.1155/2016/1984198

Barutcu, A., Briasco, E., Moon, J., Stensel, D. J., King, J. A., Witcomb, G. L., & James, L. J. (2021). Planned morning aerobic exercise in a fasted state increases energy intake in the preceding 24 h. European Journal of Nutrition, *60*(6), 3387–3396. https://doi.org/10.1007/s00394-021-02501-7

Borg, G. A. (1982). Psychophysical bases of perceived exertion. Medicine & Science in Sports & Exercise, 14(5), 377–381. https://doi.org/10.1249/00005768-198205000-00012

Buffenstein, R., Poppitt, S. D., McDevitt, R. M., & Prentice, A. M. (1995). Food intake and the menstrual cycle: a retrospective analysis, with implications for appetite research. Physiology & Behavior, 58(6), 1067–1077. https://doi.org/10.1016/0031-9384(95)02003-9

Cerin, E., Leslie, E., Sugiyama, T., & Owen, N. (2010). Perceived barriers to leisure-time physical activity in adults: an ecological perspective. Journal of Physical Activity and Health, 7(4), 451–459. https://doi.org/10.1123/jpah.7.4.451

Clayton, D. J., & James, L. J. (2016). The effect of breakfast on appetite regulation, energy balance and exercise performance. Proceedings of the Nutrition Society, 75(3), 319–327. https://doi.org/10.1017/S0029665115004243

Clayton, D. J., Barutcu, A., Machin, C., Stensel, D. J., & James, L. J. (2015). Effect of breakfast omission on energy intake and evening exercise performance. Medicine & Science in Sports & Exercise, 47(12), 2645–2652. http://doi.org/10.1249/MSS.0000000000000702

Cohen, J. (1988). Statistical power analysis for the behavioural sciences (2nd ed.). Routledge Academic.

Donnelly, J. E., Blair, S. N., Jakicic, J. M., Manore, M. M., Rankin, J. W., & Smith, B. K. (2009). American College of Sports Medicine Position Stand. Appropriate physical activity intervention strategies for weight loss and prevention of weight regain for adults. *Medicine & Science in Sports & Exercise*, *41*(2), 459–471. https://doi.org/10.1249/MSS.0b013e3181949333

Dorling, J., Broom, D. R., Burns, S. F., Clayton, D. J., Deighton, K., James, L. J., King, J. A., Miyashita, M., Thackray, A. E., Batterham, R. L., & Stensel, D. J. (2018). Acute and chronic effects of exercise on appetite, energy intake, and appetite-related hormones: the modulating effect of adiposity, sex, and habitual physical activity. Nutrients, 10(9), 1140. https://doi.org/10.3390/nu10091140

Durnin, J. V., & Womersley, J. V. G. A. (1974). Body fat assessed from total body density and its estimation from skinfold thickness: measurements on 481 men and women aged from 16 to 72 years. British Journal of Nutrition, 32(1), 77–97. https://doi.org/10.1079/BJN19740060

Edinburgh, R. M., Hengist, A., Smith, H. A., Travers, R. L., Betts, J. A., Thompson, D., Walhin, J-P., Wallis, G. A., Hamilton, L. D., Stevenson, E. J., Tipton, K. D., & Gonzalez, J. T. (2019). Skipping breakfast before exercise creates a more negative 24-hour energy balance: A randomized controlled trial in healthy physically active young men. The Journal of Nutrition, 149(8), 1326–1334. https://doi.org/10.1093/jn/nxz018

Foulds, H. J., Bredin, S. S., Charlesworth, S. A., Ivey, A. C., & Warburton, D. E. (2014). Exercise volume and intensity: a dose–response relationship with health benefits. European Journal of Applied Physiology, 114(8), 1563–1571. https://doi.org/10.1007/s00421-014-2887-9

Frampton, J., Edinburgh, R. M., Ogden, H. B., Gonzalez, J. T., & Chambers, E. S. (2022). The acute effect of fasted exercise on energy intake, energy expenditure, subjective hunger and gastrointestinal hormone release compared to fed exercise in healthy individuals: a systematic review and network meta-analysis. International Journal of Obesity, 46(2), 255–268. https://doi.org/10.1038/s41366-021-00993-1

Galloway, S. D., Lott, M. J., & Toulouse, L. C. (2014). Preexercise carbohydrate feeding and high-intensity exercise capacity: Effects of timing of intake and carbohydrate concentration. International Journal of Sport Nutrition and Exercise Metabolism, 24(3), 258–266. https://doi.org/10.1123/ijsnem.2013-0119

Gonzalez, J. T., Veasey, R. C., Rumbold, P. L., & Stevenson, E. J. (2013). Breakfast and exercise contingently affect postprandial metabolism and energy balance in physically active males. British Journal of Nutrition, 110(4), 721–732. https://doi.org/10.1017/S0007114512005582

Gonzalez, J. T., Richardson, J. D., Chowdhury, E. A., Koumanov, F., Holman, G. D., Cooper, S., Thompson, D., Tsintzas, K., & Betts, J. A. (2018). Molecular adaptations of adipose tissue to 6 weeks of morning fasting vs. daily breakfast consumption in lean and obese adults. The Journal of Physiology, 596(4), 609–622. https://doi.org/10.1113/JP275576

Griffiths, A., Deighton, K., Shannon, O. M., Boos, C., Rowe, J., Matu, J., King, R., & O'Hara, J. P. (2020). Appetite and energy intake responses to breakfast consumption and carbohydrate supplementation in hypoxia. Appetite, 147, 104564. https://doi.org/10.1016/j.appet.2019.104564

Hill, J. O., Wyatt, H. R., & Peters, J. C. (2012). Energy balance and obesity. Circulation, 126(1), 126–132. https://doi.org/10.1161/CIRCULATIONAHA.111.087213

James, R., James, L. J., & Clayton, D. J. (2020). Anticipation of 24 h severe energy restriction increases energy intake and reduces physical activity energy expenditure in the prior 24 h, in healthy males. Appetite, 152, 104719. https://doi.org/10.1016/j.appet.2020.104719

Jeukendrup, A. E., & Wallis, G. A. (2005). Measurement of substrate oxidation during exercise by means of gas exchange measurements. International Journal of Sports Medicine, 26, S28–S37. https://doi.org/10.1055/s-2004-830512

King, N. A., Hopkins, M., Caudwell, P., Stubbs, R. J., & Blundell, J. E. (2008). Individual variability following 12 weeks of supervised exercise: identification and characterization of compensation for exercise-induced weight loss. International Journal of Obesity, 32(1), 177–184. https://doi.org/10.1038/sj.ijo.0803712

Maraki, M., Tsofliou, F., Pitsiladis, Y. P., Malkova, D., Mutrie, N., & Higgins, S. (2005). Acute effects of a single exercise class on appetite, energy intake and mood. Is there a time of day effect? Appetite, 45(3), 272–278. https://doi.org/10.1016/j.appet.2005.07.005

Martin, C. K., Johnson, W. D., Myers, C. A., Apolzan, J. W., Earnest, C. P., Thomas, D. M., Rood, J. C., Johannsen, N. M., Tudor-Locke, C., Harris, M., & Hsia, D. S. (2019). Effect of different doses of supervised exercise on food intake, metabolism, and non-exercise physical activity: The E-MECHANIC randomized controlled trial. The American Journal of Clinical Nutrition, 110(3), 583–592. https://doi.org/10.1093/ajcn/nqz054

McIver, V. J., Mattin, L. R., Evans, G. H., & Yau, A. M. (2019a). Diurnal influences of fasted and non-fasted brisk walking on gastric emptying rate, metabolic responses, and appetite in healthy males. Appetite, 143, 104411. https://doi.org/10.1016/j.appet.2019.104411

McIver, V. J., Mattin, L. R., Evans, G. H., & Yau, A. M. (2019b). The effect of brisk walking in the fasted versus fed state on metabolic responses, gastrointestinal function, and appetite in healthy men. International Journal of Obesity, 43(9), 1691–1700. https://doi.org/10.1038/s41366-018-0215-x

Mears, S. A., Dickinson, K., Bergin-Taylor, K., Dee, R., Kay, J., & James, L. J. (2018). Perception of breakfast ingestion enhances high-intensity cycling performance. International Journal of Sports Physiology and Performance, 13(4), 504–509. https://doi.org/10.1123/ijspp.2017-0318

Mifflin, M. D., St Jeor, S. T., Hill, L. A., Scott, B. J., Daugherty, S. A., & Koh, Y. O. (1990). A new predictive equation for resting energy expenditure in healthy individuals. The American Journal of Clinical Nutrition, 51(2), 241–247. https://doi.org/10.1093/ajcn/51.2.241

Moholdt, T., Parr, E. B., Devlin, B. L., Debik, J., Giskeødegård, G., & Hawley, J. A. (2021). The effect of morning vs evening exercise training on glycaemic control and serum metabolites in overweight/obese men: a randomised trial. Diabetologia, 64(9), 2061–2076. https://doi.org/10.1007/s00125-021-05477-5

Monnier, L., Schlienger, J. L., Colette, C., & Bonnet, F. (2021). The obesity treatment dilemma: Why dieting is both the answer and the problem? A mechanistic overview. Diabetes & Metabolism, 47(3), 101192. https://doi.org/10.1016/j.diabet.2020.09.002

Montain, S. J., Hopper, M. K., Coggan, A. R., & Coyle, E. F. (1991). Exercise metabolism at different time intervals after a meal. Journal of Applied Physiology, 70(2), 882–888. https://doi.org/10.1152/jappl.1991.70.2.882

Naharudin, M. N., Adams, J., Richardson, H., Thomson, T., Oxinou, C., Marshall, C., Clayton, D. J., Mears, S. A., Yusof, A., Hulston, C. J., & James, L. J. (2020). Viscous placebo and carbohydrate breakfasts similarly decrease appetite and increase resistance exercise performance compared with a control breakfast in trained males. British Journal of Nutrition, 124(2), 232–240. https://10.1017/S0007114520001002

Naharudin, M. N., Yusof, A., Clayton, D. J., & James, L. J. (2021). Starving Your Performance? Reduced Preexercise Hunger Increases Resistance Exercise Performance. International Journal of Sports Physiology & Performance, 17(3), 458–464. https://doi.org/10.1123/ijspp.2021-0166

Raedeke, T. D. (2007). The relationship between enjoyment and affective responses to exercise. Journal of Applied Sport Psychology, 19(1), 105–115. https://doi.org/10.1080/10413200601113638

Robinson, S. L., Hattersley, J., Frost, G. S., Chambers, E. S., & Wallis, G. A. (2015). Maximal fat oxidation during exercise is positively associated with 24-hour fat oxidation and insulin sensitivity in young, healthy men. Journal of Applied Physiology, 118(11), 1415–1422. https://doi.org/10.1152/japplphysiol.00058.2015

Smith, H. A., & Betts, J. A. (2022). Nutrient timing and metabolic regulation. The Journal of Physiology, 600(6), 1299–1312. https://doi.org/10.1113/JP280756

Stunkard, A. J., & Messick, S. (1985). The three-factor eating questionnaire to measure dietary restraint, disinhibition and hunger. Journal of Psychosomatic Research, 29(1), 71–83. https://doi.org/10.1016/0022-3999(85)90010-8

Thompson, D., Peacock, O. J., & Betts, J. A. (2014). Substitution and compensation erode the energy deficit from exercise interventions. Medicine & Science in Sports & Exercise, 46(2), 423–423. https://doi.org/10.1249/mss.0000000000000164

Watson, D., Clark, L. A., & Tellegen, A. (1988). Development and validation of brief measures of positive and negative affect: the PANAS scales. Journal of Personality and Social Psychology, 54(6), 1063–1070. https://doi.org/10.1037/0022-3514.54.6.1063

Tables

Table 1. Participant baseline characteristics

Characteristic	Overall (n = 16)	Males (n = 8)	Females (n = 8)
Age (y)	25 ± 3	25 ± 2	24 ± 4
Weight (kg)	70.9 ± 12.1	80.6 ± 8.3	61.2 ± 4.9
Height (m)	1.74 ± 0.11	1.83 ± 0.06	1.65 ± 0.05
BMI (kg·m^{-2})	23.3 ± 1.9	24.1 ± 2.0	22.6 ± 1.6
Body fat (%)	20 ± 7	14 ± 3	26 ± 3
VO_{2peak} (mL·kg^{-1}·min^{-1})	39 ± 6	43 ± 6	36 ± 5
Dietary restraint[a]	8 ± 3	6 ± 3	9 ± 3
Dietary disinhibition[a]	5 ± 3	5 ± 3	5 ± 3
Hunger[a]	5 ± 3	5 ± 3	4 ± 2
Estimated resting metabolic rate (kcal·day^{-1})[b]	1557 ± 265	1754 ± 237	1395 ± 77

Values are means ± SD

[a] Three-factor eating questionnaire (Stunkard and Messick, 1985)

[b] Estimated via predictive equation (Mifflin et al., 1990)

Table 2. Macronutrient composition of each meal

	Carbohydrate (g)	Protein (g)	Fat (g)	Fibre (g)	Energy (kcal)
Breakfast					
FAST	93.2 ± 15.7	14.5 ± 1.0	11.2 ± 1.9	5.5 ± 0.9	543 ± 86
FED					
Lunch					
FAST	72.5 ± 11.1	36.8 ± 6.9	41.0 ± 6.2	4.1 ± 0.6	814 ± 129
FED					
Pre-Exercise Meal					
FAST	0	0	0	0	0
FED	93.2 ± 15.7	14.5 ± 1.0	11.2 ± 1.9	5.5 ± 0.9	543 ± 86
***Ad-Libitum* Post-Exercise Meal**					
FAST	152.1 ± 60.3	23.9 ± 9.5	17.9 ± 7.2	8.4 ± 3.3	882 ± 350*
FED	135.0 ± 48.4	21.2 ± 7.6	15.9 ± 5.7	7.4 ± 2.7	783 ± 281
Total					
FAST	317.7 ± 82.4	75.3 ± 16.3	70.1 ± 14.4	18.0 ± 4.6	2239 ± 533*
FED	393.8 ± 80.9	87.1 ± 14.6	79.3 ± 14.3	22.5 ± 4.5	2682 ± 519

Data are mean ± SD

[*] Values are significantly different from FED ($P < 0.05$).

Table 3. Pre- and Post-Exercise Subjective Responses

	FAST	FED
PANAS Positive Affect[a]	22 ± 6*	26 ± 6
PANAS Negative Affect[a]	13 ± 3	12 ± 3
PACES-8 Score[b] (%)	49 ± 12*	57 ± 13

Values are means ± SD

* Values are significantly different from FED ($P < 0.05$).

[a] PANAS questionnaire (Watson et al., 1988)

[b] PACES-8 questionnaire (Raedeke, 2007)

Figures

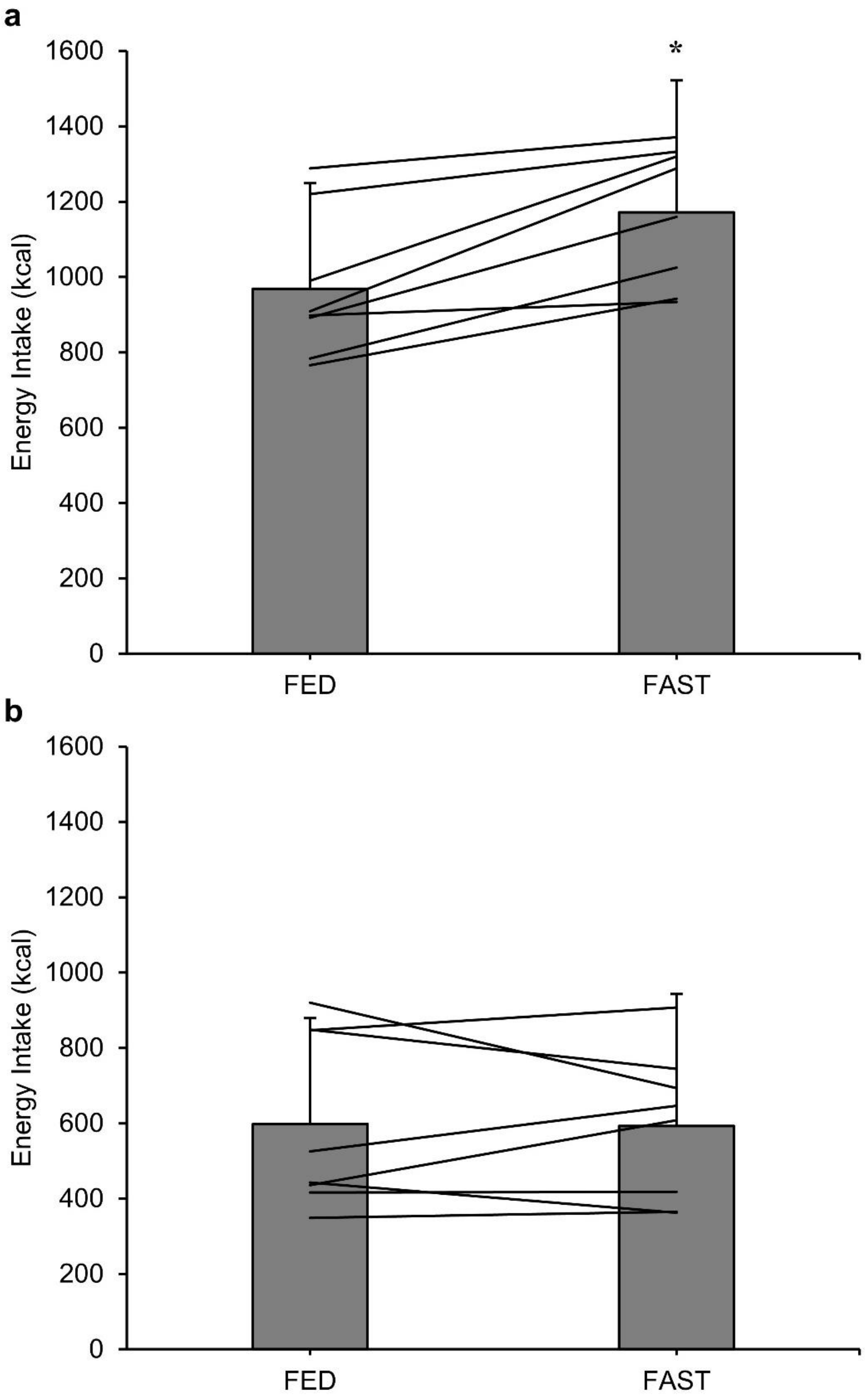

Figure 1. Energy intake (kcal) at the *ad-libitum* meal for **(a)** males (*n*=8), and **(b)** females (*n*=8). The bars display mean values, with vertical error bars representing SD. The lines display individual participants' *ad-libitum* energy intake for each experimental trial. * Significant difference between FED and FAST ($P < 0.05$).

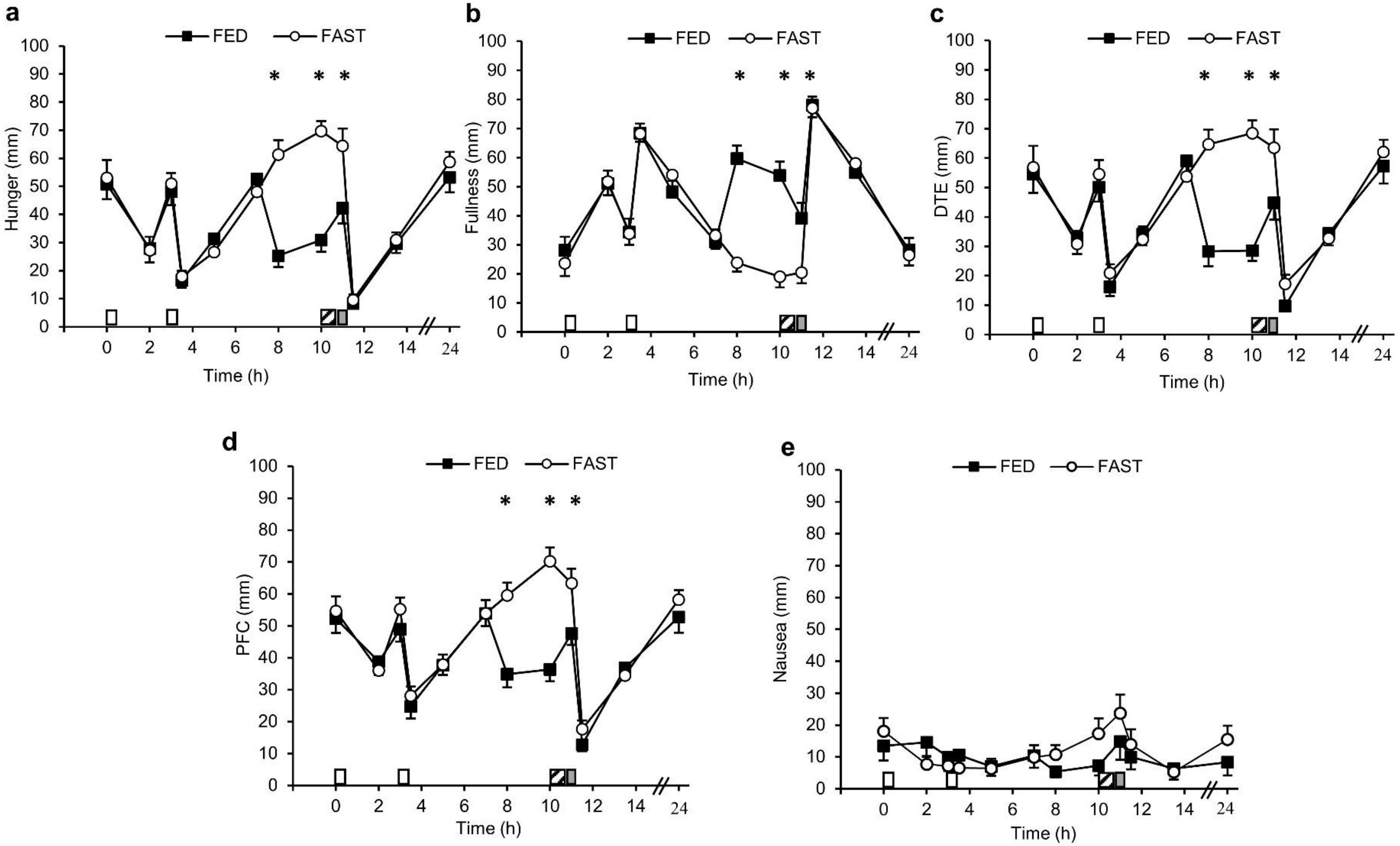

Figure 2. (a) Hunger, **(b)** fullness, **(c)** desire to eat (DTE), **(d)** prospective food consumption (PFC), and **(e)** nausea in FED and FAST. Data are mean ± SEM. White rectangles represent standardised meals; grey rectangle represents *ad-libitum* meal; diagonal striped rectangle represents exercise. * Significant difference between FED and FAST ($P < 0.05$).

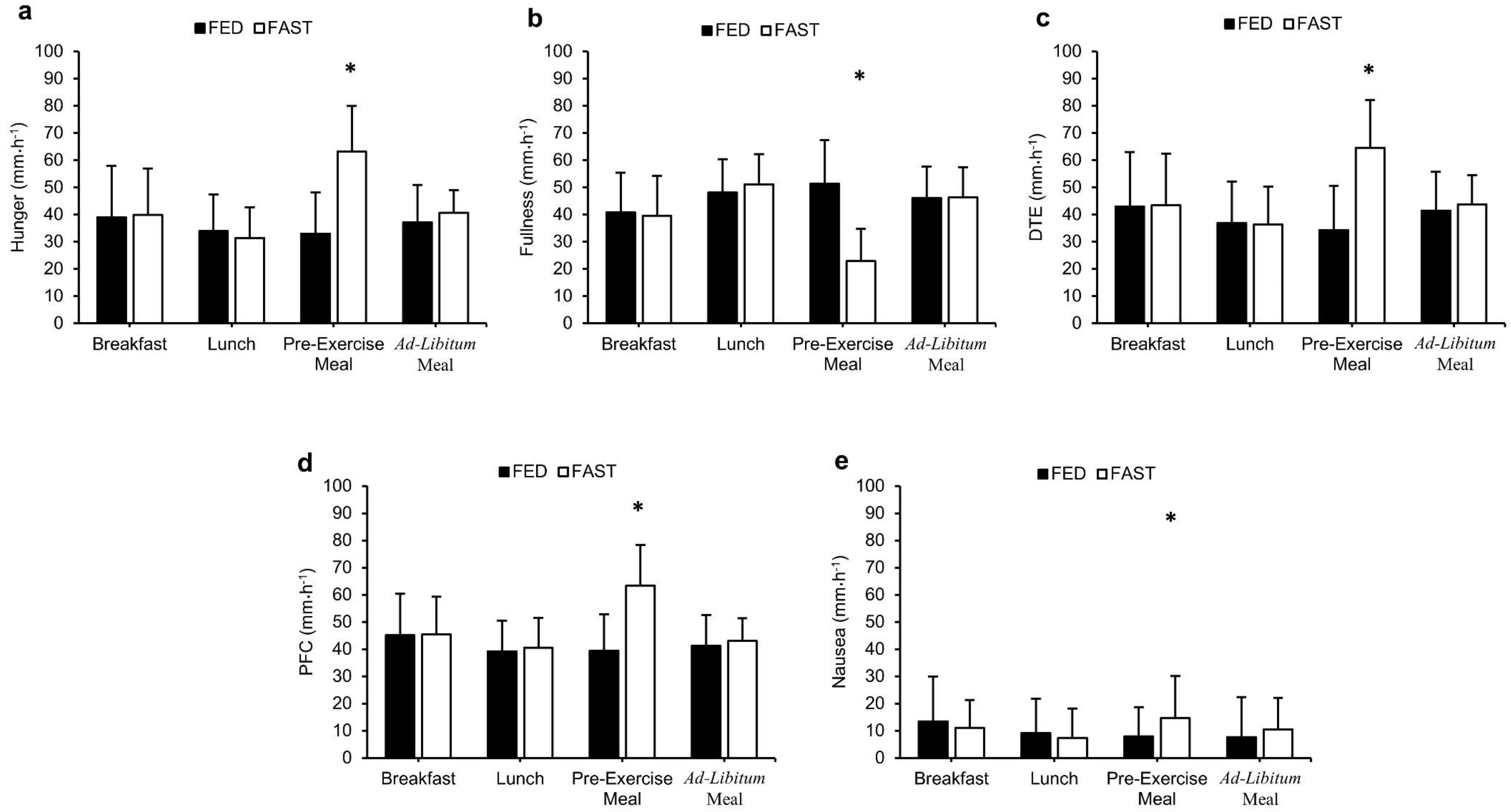

Figure 3. **(a)** Hunger, **(b)** fullness**, (c)** desire to eat (DTE), **(d)** prospective food consumption (PFC), and **(e)** nausea time-averaged area under the curve (AUC) in FED and FAST. Data are mean ± SEM. * Significant difference between FED and FAST ($P < 0.05$)

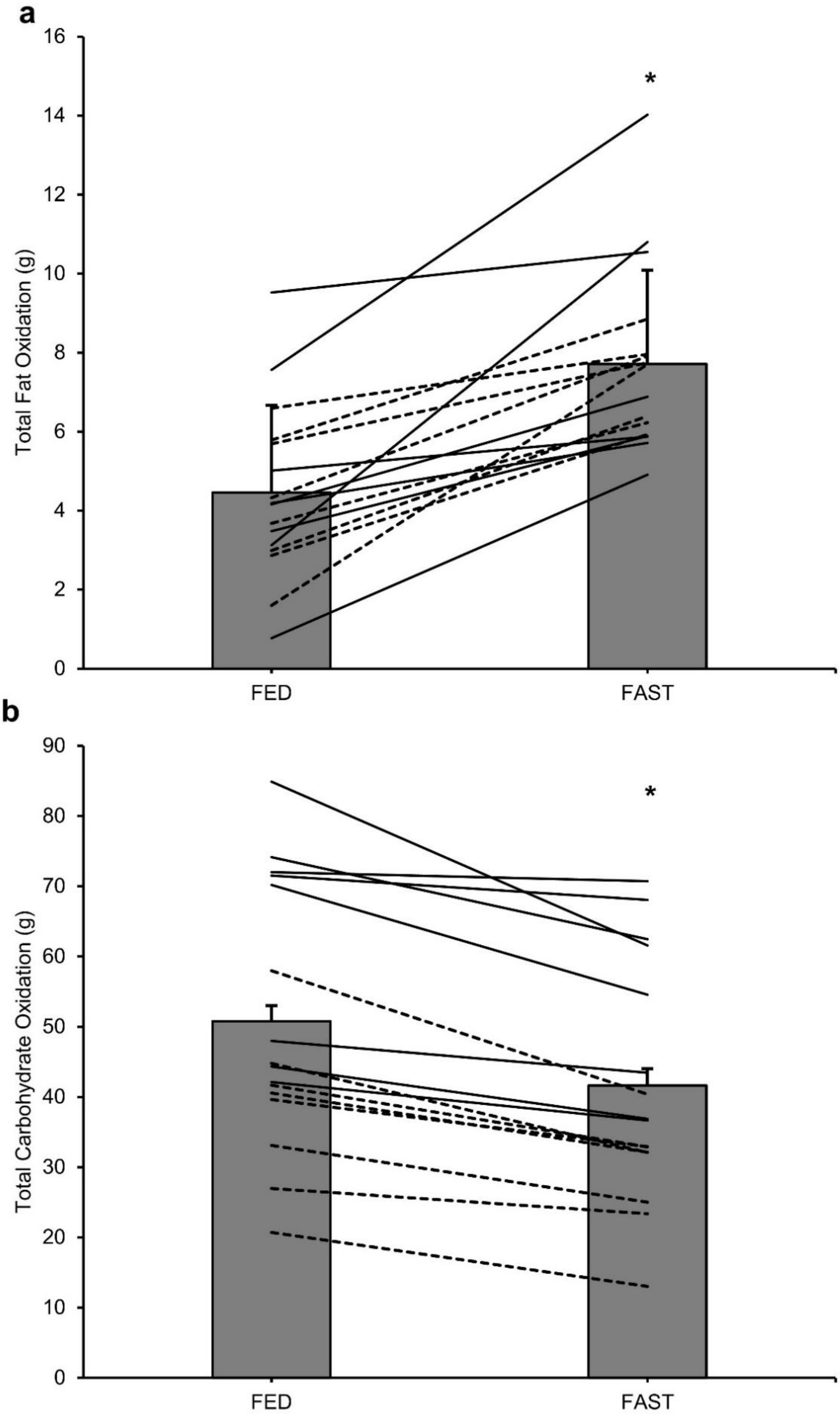

Figure 4. (a) Total fat oxidation (g), and **(b)** total carbohydrate oxidation (g) during the 30-min steady-state bout of cycling in FED and FAST. Data are mean ± SD. The lines display individual participants' substrate oxidation during each experimental trial (dotted line: females; block line: males). * Significant difference between FED and FAST ($P < 0.05$).

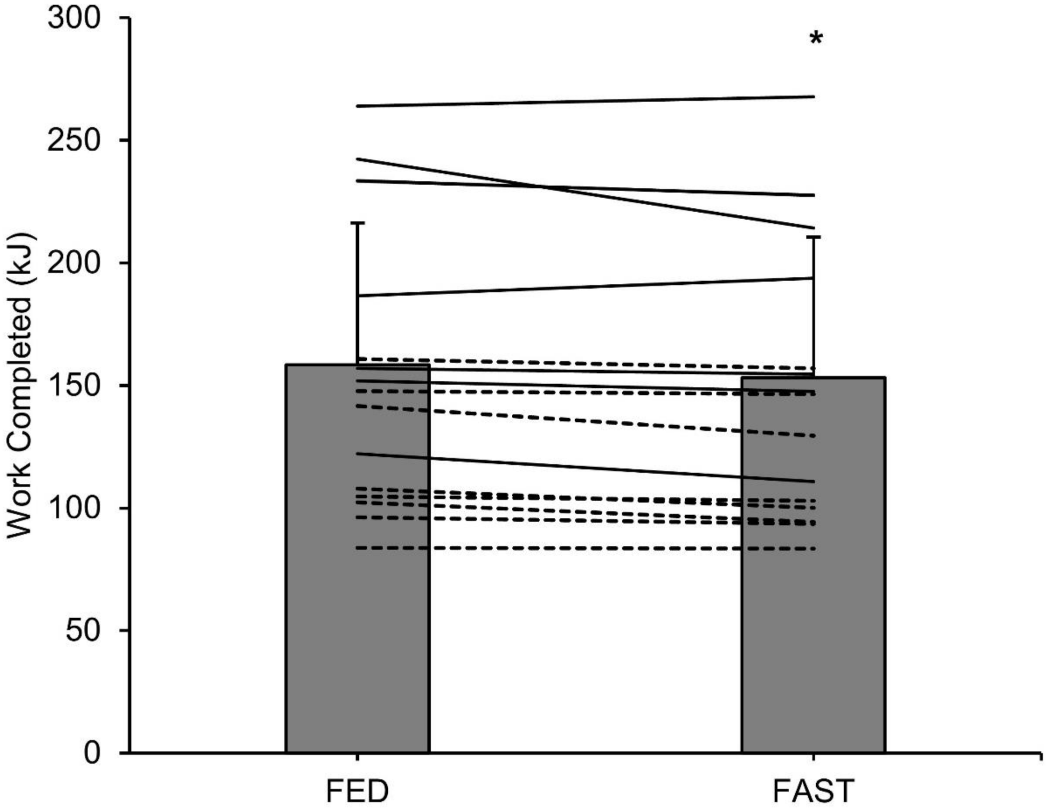

Figure 5. Total work completed (kJ) during the 15-min exercise performance test in FED and FAST. Data are mean ± SD. The lines display individual participants’ completed work during each experimental trial (dotted line: females; block line: males). * Significant difference between FED and FAST ($P < 0.05$).