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Effects of acute caffeine supplementation on reducing exercise-associated hypoglycaemia in individuals with Type 1 diabetes mellitus

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Abstract

Aim To determine the effects of acute caffeine ingestion on glycaemia during moderate to vigorous intensity aerobic exercise and in recovery in individuals with Type 1 diabetes.

Methods A total of 13 patients with Type 1 diabetes [eight women, five men: mean ± SD age 25.9 ± 8.8 years, BMI 71.9 ± 11.0 kg, maximal oxygen consumption 46.6 ± 12.7 ml/kg/min, body fat 19.9 ± 7.2%, duration of diabetes 14.4 ± 10.1 years and HbA_{1c} 55 ± 8 mmol/mol (7.4 ± 0.8%)] were recruited. Participants ingested capsules that contained gelatin or pure caffeine (6.0 mg/kg body mass) and performed afternoon exercise for 45 min at 60% maximal oxygen consumption on two separate visits with only circulating basal insulin levels.

Results The main finding was that a single caffeine dose attenuates the drop in glycaemia by 1.8 ± 2.8 mmol/l compared with placebo intake during exercise (P=0.056). Continuous glucose monitoring data, however, showed that caffeine was associated with elevated glycaemia at bedtime after exercise, compared with placebo, but lower glucose concentrations in the early morning the next day.

Conclusions Caffeine intake should be considered as another strategy that may modestly attenuate hypoglycaemia in individuals with Type 1 diabetes during exercise, but should be taken with precautionary measures as it may increase the risk of late-onset hypoglycaemia.

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Introduction

Regular moderate to vigorous intensity exercise is associated with numerous psychological and physiological benefits for individuals with Type 1 diabetes [1]. Some of the benefits of habitual exercise include potentially lowering HbA_{1c} in individuals with Type 1 diabetes, increasing cardiovascular and aerobic fitness, increasing whole-body insulin sensitivity, and improving overall quality of life [2]. Despite the known benefits of habitual exercise, however, a critical risk that must be considered for individuals with Type 1 diabetes is hypoglycaemia [2]. One of the largest barriers to exercise participation for individuals with Type 1 diabetes is the fear of hypoglycaemia during the activity and in recovery [3].

There are various strategies that individuals with Type 1 diabetes may adopt before, during and after exercise to mitigate the likelihood of hypoglycaemia [4]. Before exercising, it is important to determine the time, duration and intensity of the activity so that the proper adjustments to insulin ratios and food intake can take place. For example, the premeal insulin dose can be reduced anywhere from 25 to 75%, depending on the intensity and duration of the activity [5]. Another common strategy to avoid hypoglycaemia during exercise is the consumption of fast-acting carbohydrates [2]; however, none of these strategies provides complete assurance that hypoglycaemia will not occur, and these strategies require knowledge of insulin pharmacokinetics and advanced planning.

Caffeine (1,3,7-trimethylxanthine) is the most commonly consumed chemical stimulant in the world [6]. The ingestion of caffeine has numerous effects throughout the body including on the gastrointestinal, cardiovascular, central nervous, metabolic, renal, and respiratory systems [7]. Caffeine causes an increase in lipolysis in adipose tissues and hepatic glucose production in the liver, with a decrease in glucose uptake in skeletal muscle [7]. Although numerous studies have focused on the effects of caffeine on metabolism

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What's new?

- We examined the role of caffeine ingestion on exercise-associated hypoglycaemia in individuals with Type 1 diabetes.
- We found that caffeine intake increased hypoglycaemia awareness and elevated blood glucose concentrations during 45-min exercise in patients with diabetes.
- We also found that a single caffeine dose did not protect against nocturnal hypoglycaemia in the early-morning hours and may, in fact, increase the risk.

during exercise [8], there is a lack of research specifically on caffeine, exercise and Type 1 diabetes.

Caffeine's potential to increase blood glucose levels during exercise by increasing glucose production and/or restraining glucose disappearance may have benefits for the prevention of hypoglycaemia in individuals with Type 1 diabetes. Caffeine intake in Type 1 diabetes has been reported to lower nocturnal hypoglycaemia risk [9] and enhance hypoglycaemia awareness [10]. Recently, one pilot study in five patients with Type 1 diabetes (all receiving multiple daily injection treatment) found that acute caffeine consumption (5.0 mg/kg body mass) reduced the likelihood of hypoglycaemia during exercise [11]. In the present study, we examined the effects of acute caffeine supplementation on blood glucose concentrations and energy metabolism during exercise and on glycaemia after exercise in patients with Type 1 diabetes.

Patients and methods

Study participants

The experimental protocol conformed to the standards set by the Declaration of Helsinki and is approved by the Research Ethics Board at York University. A total of 13 patients (five men, eight women) with Type 1 diabetes were recruited for this study by word of mouth in the Toronto area. The inclusion criteria stipulated that participants must have had diabetes for > 1 year, treated either with continuous subcutaneous insulin infusion or multiple daily injection therapy, and maintain fair-to-good glycaemic control (last $HbA_{1c} \leq 75$ mmol/mol; 9.0%) [12]. Based on a self-reported questionnaire, three of the participants were classified as non-users of caffeine and 10 as caffeine users. Participants were also screened for any cardiovascular complications and/or contraindications to aerobic exercise using the Physical Activity Readiness Questionnaire for Everyone [13].

Experimental design

A double-blind, placebo-controlled crossover design was used which included a total of three laboratory visits. The first visit was a protocol familiarization and determination of maximal aerobic power [maximal oxygen consumption (VO_{2max})], followed by two experimental visits during which the prolonged moderate to vigorous intensity exercise sessions, either with or without caffeine supplementation, took place. All of the participants were asked to avoid caffeine-containing beverages and alcohol consumption and to refrain from exercise 24 h before each visit.

Fitness assessment (visit 1)

The initial visit was a familiarization session, during which anthropometric measurements including height, body mass and body fat percentage were measured. VO_{2max} and maximum heart rate were measured using an incremental-to-maximum effort treadmill protocol. Expired air was collected from a corrugated plastic hose into a 120-litre Tissot gasometer (Warren E. Collins Ltd., Braintree, MA, USA) and gas composition was measured using rapid response electrochemical analysers (AEI Technologies, Pittsburgh, PA, USA). After a 2–3-min break, participants were asked to complete a supramaximal workload assessment with increasing slope to confirm the attainment of VO_{2max} [14]. VO_{2max} was confirmed when oxygen consumption (VO_2) reached a plateau (within 150 ml/min of the previous workload) or decreased with progressively increasing workloads, along with at least one of the following criteria: respiratory exchange ratio > 1.0, achieved age-predicted maximum heart rate or volitional fatigue [15].

Prolonged exercise sessions (visits 2 and 3)

During visit 2, participants arrived at the Human Performance Laboratory at York University and the iPro2 (Medtronic Ltd, Minneapolis, MN, USA) continuous glucose monitor (retrospective, non real-time analysis) was inserted into the interstitial space under the skin on the abdomen or upper buttocks using a Sen-Serter (Medtronic Ltd) according to the manufacturer's instructions ~4 h before exercise. Participants wore the same continuous glucose monitor for the duration of visits 2 and 3 (within 4–6 days) and were instructed to use the provided glucometer for all self-monitoring of blood glucose. A standardized lunch (within subject) was consumed and a bolus of insulin was taken no later than 12:00 h for all participants during visit 2 and 3, so the amount of 'active' on-board prandial insulin from lunchtime would be minimal at the start of exercise (16:00 h).

On arrival at the laboratory at 15:00 h, participants were instructed to ingest the capsules (caffeine or placebo) provided by the investigator who was also blinded to the treatment. The capsules contained 6.0 mg/kg of body weight of either pure caffeine (Sigma-Aldrich, St. Louis, MO, USA) or 6.0 mg/kg of gelatin (placebo). A catheter (SURFLO Teflon IV Catheter, Somerset, NJ, USA) was inserted into the

antecubital vein and 4.0 ml heparinized vacutainers were used to collect blood at each time point (baseline, 20 min into exercise and post-exercise). Ten minutes before exercise, all participants ingested 16 g of dextrose (Dex4©, AMG Medical Inc., Montreal, Canada). Participants exercised at 60–70% of VO_{2max} for 45 min, and capillary glucose levels were measured at 15-min intervals. Visits were rescheduled if glucose concentrations were ≥ 16.0 mmol/l before the onset of exercise and were terminated immediately if glucose levels reached 3.9 mmol/l during exercise.

Substrate oxidation

Expired gas was analysed during the initial and final 10 min of both visits to measure oxygen consumption and carbon dioxide production and substrate utilization. For this, expired air was analysed, using AEI analysers (see above) for the fractional concentration of oxygen (F_EO_2) and carbon dioxide (F_ECO_2) immediately after the completion of each gas collection (Models S-3A and CD-3S, respectively; Applied Electrochemistry, Sunnyvale, CA, USA). The O_2 and CO_2 analysers were calibrated frequently with gases of known concentrations (Praxair, Toronto, ON, Canada). Simultaneous measurements of the gas temperature and atmospheric pressure were made to correct the measured gas volumes to a standard temperature and pressure. Absolute VO_2 (l/min) values were calculated using the corrected volume of expired air together with F_EO_2 and F_ECO_2. Fat and carbohydrate oxidation rates were determined using standard equations, which assume negligible contribution from protein oxidation [16].

Blood samples

Whole-blood samples were drawn at baseline, 20 min into exercise, and immediately after exercise in a subset of participants (n=11). Samples were left at room temperature for 5 min and then centrifuged at 4000 × g for 8 min. Plasma was extracted from vacutainers and 250 µl was aliquoted into five 1.0-ml tubes. Each tube had a measured concentration of EDTA:trasylol solution, where 2.4 g of EDTA (Sigma-Aldrich Canada Ltd., Oakville, Canada) was dissolved in 100 ml of double distilled water on a hot plate and trasylol (Bayer Pharmaceuticals, Toronto, Canada) preservative was added in a 1:1 ratio to the EDTA solution. Once plasma was equally distributed among tubes, samples were spun using a Labnet VX100 vortex (Mo Bio Laboratories, Carlsbad, CA, USA) and immediately frozen in a -80°C freezer.

Plasma samples, performed in duplicate, were used to measure non-esterified free fatty acids (NEFA; Wako Pur Chemical Industries, Osaka, Japan) and adrenaline concentrations (BA-E 6100 Rocky Mountain Diagnostics, Colorado Springs, CO, USA). The adrenaline interassay plasma coefficient of variation, as reported by the manufacturer was 14.5%. Plasma glucose concentrations were measured using a Yellow Springs Instrument glucose oxidase analyser (YSI 2300 Stat Plus Glucose and L-Lactate Analyser; YSI, Yellow Springs, OH, USA) to determine the correlation between capillary whole-blood glucose values and plasma glucose levels. A 250-µl plasma sample was used to measure glucose concentrations.

Statistics

Glucose concentrations during the pre-exercise and exercise periods were analysed separately using two-way (time by trial) repeated measures ANOVA. Percent time spent in different glucose concentration zones (i.e. hypoglycaemia, euglycaemia, hyperglycaemia) from 18:00 h to 06:00 h after exercise were compared between visits using the Mann–Whitney test for non-parametric statistics. For this analysis, euglycaemia was defined as a continous glucose monitor-assessed glucose concentration of 4.0–9.9 mmol/l; hypoglycaemia as ≤ 3.9 mmol/l; and hyperglycaemia as ≥ 10.0 mmol/l. Circulating adrenaline and NEFA levels were also analysed using two-way (time by trial) repeated measures ANOVA. Tukey's honest significant difference *post hoc* test was used if significant interactions were found. Statistical significance was set at P <0.05 unless otherwise indicated. Participants' descriptive characteristics were reported using mean ± SD values and are shown in Table 1. All statistical analyses were conducted using STATISTICA 7.0 (StatSoft, Tulsa, OK, USA) and GRAPHPAD PRISM software (Version 4.0).

Results

Study participants

A total of 13 participants with Type 1 diabetes were included in the final analysis (five men and eight women).

Table 1 Plasma glucose, free fatty acid and adrenaline levels during the caffeine and placebo visits

	Before exercise	During exercise	After exercise
Plasma glucose, mmol/l			
Caffeine	9.5 ± 2.8	10.8 ± 3.0	8.4 ± 3.5
Placebo	8.1 ± 1.9	8.5 ± 3.0	5.6 ± 3.0
Adrenaline, pg/ml			
Caffeine	41.3 ± 16.1		61.4 ± 12.9
Placebo	30.4 ± 9.3		51.9 ± 15.9
NEFA, mmol/l			
Caffeine	0.35 ± 0.35	0.28 ± 0.29	0.76 ± 0.73
Placebo	0.20 ± 0.22	0.24 ± 0.18	0.61 ± 0.53

NEFA, non-esterified free fatty acids.
Data are mean ± SD.
Compared with the placebo visit, the caffeine visit was associated with higher plasma glucose concentrations (main effect of visit; P=0.03) and higher adrenaline concentrations (main effect of visit; P=0.01). Free fatty acid levels increased with exercise (P=0.01 for main effect of time) but did not differ significantly between visits. No significant interaction between visit and time existed in these analyses.

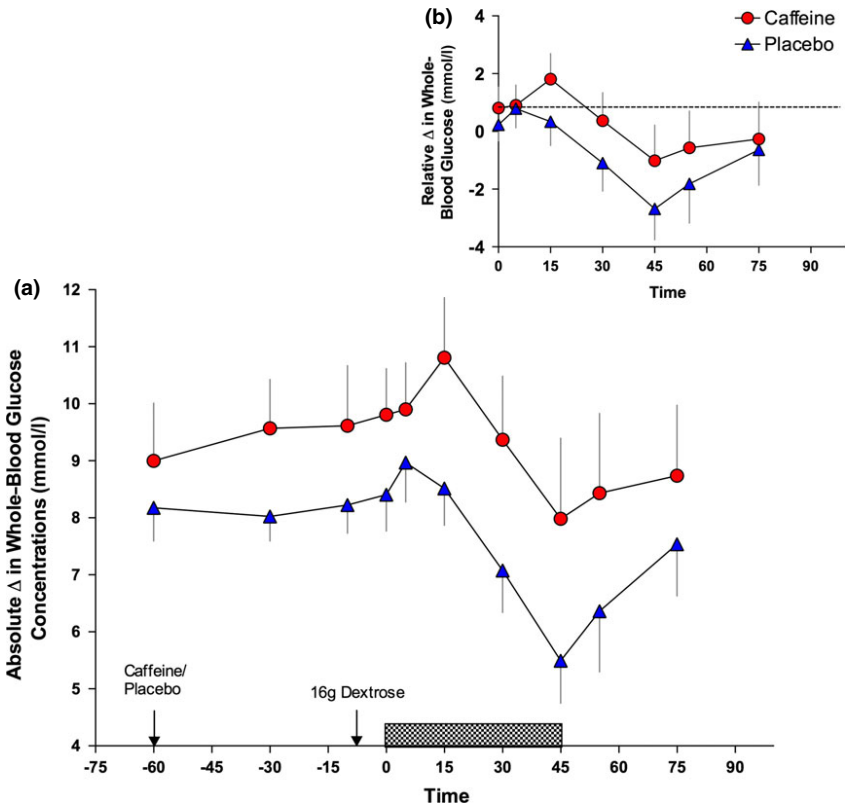

FIGURE 1 (a) Absolute and (b) relative change in whole-blood capillary glucose concentrations during the caffeine and placebo visits. Hashed box denotes the exercise protocol (45 min of treadmill running at 60–70% of maximum oxygen consumption). The caffeine trial (circles) is denoted with lower error bars removed and the placebo trial (triangles) is denoted with upper error bars removed. Two-way repeated measures ANOVA was used to measure statistical significance (n=13). Trial by time interaction approached statistical significance (P=0.079). The caffeine visit was associated with higher overall glucose levels (main effect of visit, P=0.056). No significant differences were seen in relative whole-blood glucose concentrations during exercise, between caffeine and placebo visits.

Women were not studied during any particular phase of their menstrual cycle. Participants varied in mean age (25.9 ± 8.8 years), height (172.0 ± 10.3 cm), body mass (71.9 ± 11.0 kg), VO_{2max} (46.6 ± 12.7 ml/kg/min), body fat (19.9 ± 7.2%), waist circumference (82.4 ± 4.0 cm) and duration of diabetes (14.4 ± 10.1 years). All participants' HbA_{1c} values were measured during their exercise visit 2 using an A_{1c}Now+ meter (Bayer, Berkeley, CA, USA). All of the participants had fair to good metabolic control based on the last HbA_{1c} measurement [55 ± 8 mmol/mol (7.4 ± 0.8%)]. The participants were using either continuous subcutaneous insulin infusion (n=10) or multiple daily injections (n=3). The types of insulin being used included Novorapid® (Novo Nordisk Pharmaceuticals, Inc.), Humalog® (Eli Lilly and Company) and long-acting insulin Lantus® (Sanofi Aventis) and Levemir® (Novo Nordisk Inc.). Participants varied in their self-reported exercise levels from mildly active to elite. Elite was defined as participating in a sport or activity at the national or international level. The majority of participants fell into the category of moderate- to high-level of activity and only one participant was categorized as not active.

Blood glucose response before, during and after exercise

Figure 1 shows the time-course changes in absolute and delta change (inset) in whole blood glucose concentrations during the caffeine and placebo visits. There were no significant differences in the absolute pre-exercise (baseline to time point zero) blood glucose concentration between the two visits (P=0.18). Mean ± SD blood glucose concentrations were 1.8 ± 2.8 mmol/l higher in the caffeine vs. the placebo visit during the exercise period (main effect of visit; P=0.04, trial by time interaction; P=0.079); however, caffeine intake did not appear to alter the rate of change in whole-blood glucose concentration during exercise between the 15–45-min time points. Of the 13 participants, one developed documented symptomatic hypoglycaemia (blood glucose ≤ 3.9 mmol/l) [17] during the placebo visit 30 min into exercise, and the session was terminated prematurely. By the end of exercise, six participants had glucose levels ≤ 3.9 mmol/l in the placebo visit vs. two in the caffeine visit (P=0.04, one-tailed chi-squared analysis). In early recovery, both visits were associated with similar rates of increase in glucose concentrations (Fig. 1). Table 1 shows the plasma

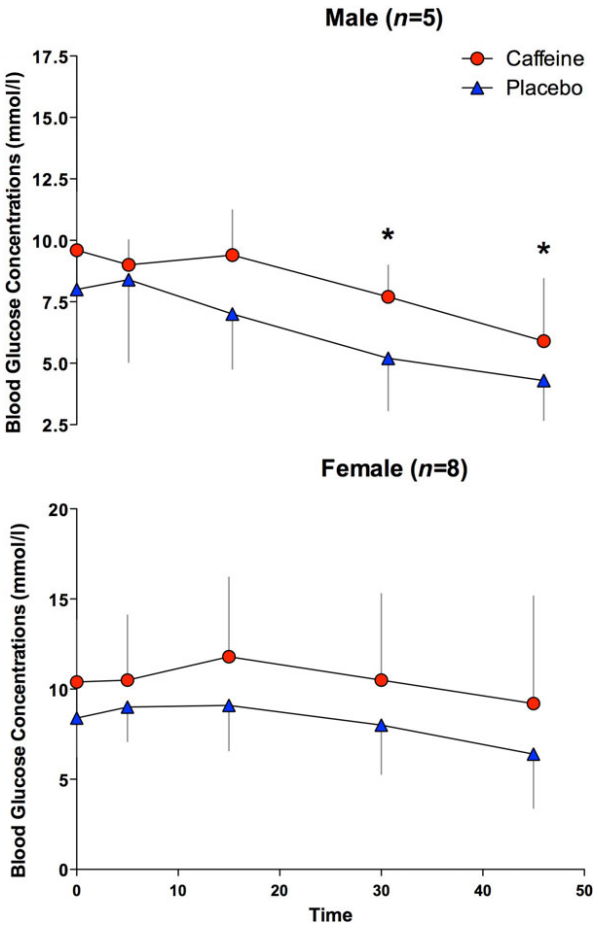

FIGURE 2 Absolute change in whole-blood capillary glucose concentrations, as measured by self-monitoring of blood glucose in men (n=5) and women (n=8) during placebo (triangles) and caffeine (circles) visits. Two-way repeated measures ANOVA was used to measure statistical significance (n=13). Men show a significant change in blood glucose concentrations with visit by time (P=0.02). Women tended to have higher blood glucose concentrations during the caffeine visit compared with the placebo visit (P=0.06). *P<0.05.

glucose values obtained intravenously using a glucose oxidase analyser before, during and after exercise. Plasma glucose values were 2.0 ± 2.6 mmol/l higher during the caffeine visit compared with the placebo visit (main effect of visit; P=0.03).

Figure 2 shows the absolute whole-blood glucose concentrations during exercise in men and women. In men, caffeine intake attenuated the decline in whole-blood glucose during exercise (trial by time interaction; P=0.02). In women, compared with placebo, caffeine was associated with a 2.3 ± 2.9 mmol/l higher glucose concentration throughout the entire exercise protocol (main effect of trial; P=0.06).

Figure 3 shows the correlation between perceived blood glucose levels during exercise and measured blood glucose levels during both placebo and caffeine visits. Participants were asked to guess their blood glucose levels during exercise (perceived glucose) and were blinded to the actual measured

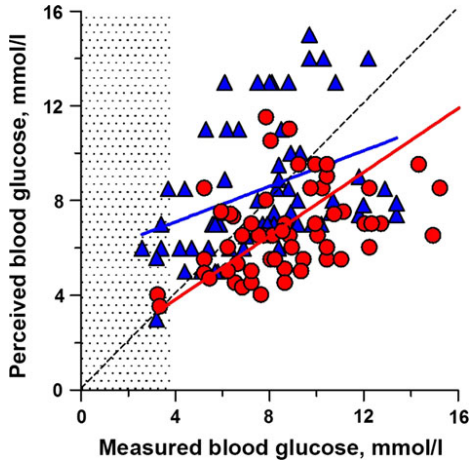

FIGURE 3 Measured versus perceived blood glucose concentrations during exercise in caffeine (circles) and placebo (triangles) visits. The dotted line represents the line of identity. There were significant correlations between measured and perceived glucose levels in both visits but the correlation was stronger in the caffeine trial (r=0.77) than in the placebo visit (r=0.36).

value until the end of exercise. There were significant correlations between measured and perceived glucose levels in both visits but the correlation was stronger in the caffeine visits (r=0.77) than in the placebo visit (r=0.36). Moreover, there was a tendency for individuals to underestimate their blood glucose levels during the caffeine visit, particularly in the hyperglycaemic range. In the placebo visit, participants had a tendency to overestimate their glucose levels, particularly in the hypoglycaemic range.

Metabolic and hormonal responses to exercise

The NEFA and adrenaline concentrations are shown in Table 1 for both placebo and caffeine visits. NEFA concentrations were similar between visits but increased significantly with time in both placebo and caffeine conditions (P=0.01). Adrenaline concentrations were similar pre-exercise between visits, but increased more with caffeine post-visit (P=0.01).

Substrate oxidation during exercise

The VO_2, respiratory exchange ratio and substrate oxidation rates during exercise are shown in Table 2. The respiratory exchange ratio values significantly decreased from the start to the end of exercise in both placebo and caffeine visits, but the drop was greater in the caffeine visit. Fat oxidation rates increased significantly in both caffeine and placebo visits from the start to the end of exercise, with a greater increase in the caffeine visit. Carbohydrate oxidation rates decreased significantly in both caffeine and placebo visits from the start to the end of exercise, with a greater decline during the caffeine visit.

Table 2 Maximum oxygen consumption, respiratory exchange ratio and substrate oxidation rates at the beginning of exercise (5–10-min time period) and at the end of exercise (40–45 min) in the placebo and caffeine visits

Visit	VO_2, l/min		Respiratory exchange ratio		Fat oxidation, g/min		Carbohydrate oxidation, g/min	
	Beginning of exercise	End of exercise	Beginning of exercise	End of exercise	Beginning of exercise	End of exercise	Beginning of exercise	End of exercise
Caffeine	2.24 ± 0.47	2.00 ± 0.77	0.96 ± 0.03	0.92 ± 0.04*†	0.17 ± 0.10	0.29 ± 0.20*†	2.65 ± 0.78	1.88 ± 0.92*†
Placebo	2.20 ± 0.45	1.92 ± 0.77	0.96 ± 0.05	0.94 ± 0.05*	0.17 ± 0.19	0.19 ± 0.16*	2.57 ± 1.13	2.04 ± 0.95*

VO_2, oxygen consumption.
Data are mean ± SD.
VO_2 had a tendency to decrease from the start to the end of exercise, but this did not reach significance. **respiratory exchange ratio** decreased significantly from the start to the end of exercise in both caffeine and placebo visits. Fat oxidation rates increased significantly from the start to the end of exercise and carbohydrate oxidation rates had the opposite effect.
*Significantly different from beginning of exercise. †Significantly different from placebo.

Evening and nocturnal glycaemia after exercise

A subset of participants (*n*=10) had overnight continuous glucose monitor data available for both visits (Fig. 4). Interstitial glucose values after the caffeine visit tended to be more stable throughout the early evening hours, while values dropped in the placebo visit. From approximately 02:30h until 06:00h, however, the glucose values dropped markedly (~4 mmol/l) in the caffeine visit, but not in the placebo visit (*P*=0.04). The number of participants with glucose concentrations ≤ 3.9 mmol/l was five in the caffeine visit vs. two in the placebo visit (*P*=0.16, chi-squared

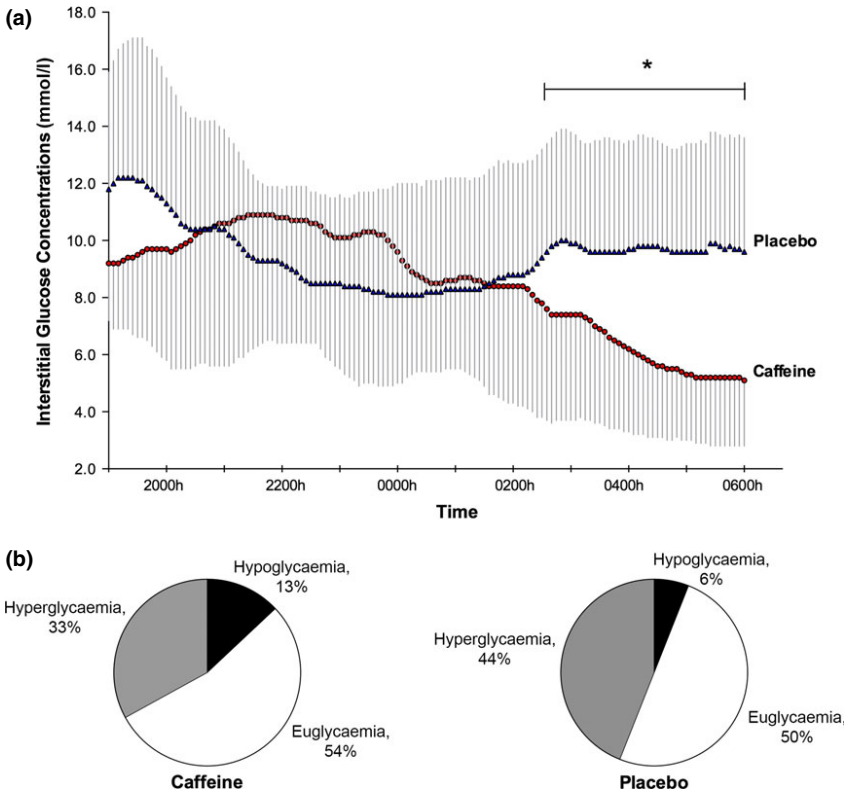

FIGURE 4 Evening and nocturnal interstitial glucose concentrations, as measured using continuous glucose monitor (CGM) (panel A), along with a pie chart representation of the percent time spent in euglycaemia, hypoglycaemia, and hyperglycaemia (panel B). Placebo trial (blue triangle) with lower error bars removed and caffeine trial (red circle) with upper error bars removed (n=10). A two-way repeated measures ANOVA design was used to determine statistical significance. There were no significant differences (*P*=0.35) in interstitial glucose concentrations in the evening hours (6PM – 11:30PM). In the early morning hours (2:30AM – 6:00AM) interstitial glucose levels were significantly lower following the caffeine visit (*P*=0.04). * denotes *P*<0.05.

analysis). The percentage of time spent in hypoglycaemia, euglycaemia and hyperglycaemia between 18:00h and 06:00h the following day is shown in Figure 4b. The percent time in hypoglycaemia tended to be higher after caffeine intake (13 ± 20%) compared with placebo intake (6 ± 12%), but this difference failed to reach statistical significance (Mann–Whitney test for non-parametric statistics, P=0.17). Percent time in euglycaemia and hyperglycaemia were not significantly different between visits (Figure 4b).

Discussion

The present study shows that a single caffeine dose of 6.0 mg/kg (equivalent to about four to six cups of coffee for an adult man) attenuated the drop in glycaemia during 45 min of steady-state aerobic exercise by 1.8 ± 2.8 mmol/l compared with placebo intake; however, continuous glucose monitor data also showed that caffeine intake was associated with elevated blood glucose concentrations at bedtime but, paradoxically, lower glucose levels during the early morning hours. These findings should temper the enthusiasm for caffeine to prevent exercise-associated hypoglycaemia in individuals with Type 1 diabetes.

Previous studies have shown that moderate to high caffeine intake (3.0–6.0 mg/kg of body mass) increases the warning signs of hypoglycaemia at rest [9] and can improve heart rate variability [18] in patients with Type 1 diabetes; however, few studies have looked at the implications on blood glucose homeostasis during and after exercise in Type 1 diabetes. Interestingly, caffeine ingestion in the present study was associated with a decrease in perceived blood glucose concentrations, as was also observed in participants at rest in a study by Richardson *et al.* [9]. As both caffeine and exercise cause increases in catecholamines, which are also released during hypoglycaemia, participants may have believed that they were developing hypoglycaemia during the caffeine visit, as has been reported previously in participants with Type 1 diabetes at rest [18].

In the present study, caffeine was associated with higher NEFA and adrenaline levels, both of which may have contributed to the higher glucose levels by reducing glucose disposal into muscle. The drop in respiratory exchange ratio was greater in the caffeine visit compared with the placebo visit and there was a great reliance on fat oxidation with caffeine ingestion (Table 2), findings that have long been associated with caffeine intake in healthy athletes [19].

Increased carbohydrate intake before, during and after exercise is one of the most common guidelines for maintaining blood glucose levels to avoid hypoglycaemia [2]; however, ingesting a caffeine supplement as an alternate strategy for reducing exercise-induced hypoglycaemia may offer some advantages for some individuals. First, caffeine consumption does not appear to promote marked hyperglycaemia in most individuals (Fig. 1, Fig. S1), as can be observed when prandial insulin to carbohydrate ratio is heavily reduced [20,21]. Second, caffeine does not increase caloric consumption, nor is it associated with gastric distress (unless consumed in excess of 9.0 mg/kg of body mass), which are common side effects of excessive carbohydrate intake [22]. In the present study, participants ingested a limited amount of carbohydrates (16 g dextrose) in both visits to protect against severe hypoglycaemia, particularly in the placebo visit. When combined with caffeine intake, this amount of carbohydrate appears effective in limiting the drop in glycaemia during 45 min of vigorous aerobic exercise, although the effectiveness of this strategy did vary markedly among participants (Fig. S1).

We found a high degree of variability in the glycaemic responses to exercise in both visits, with more variability in the caffeine visit compared with the placebo visit (Fig. S1). It is well established that not all individuals metabolize caffeine in the same way [23] and it is known that individuals that are homozygous for the cytochrome P450 1A2 (CYP1A2)*1A allele more rapidly metabolize caffeine in comparison with carriers of the CYP1A2*1F allele [23]. If blood samples were analysed for DNA isolation, it would have allowed for a differentiation between caffeine responders and non-responders in the present study. Interestingly, a sub-analysis (data not shown) showed a 4.0-mmol/l higher glucose concentration with caffeine intake in non-habitual caffeine users vs. a 1.0-mmol/l increase in glucose with caffeine users. Previous studies conducted in young healthy men (without diabetes) show that acute caffeine intake (5.0 mg/kg) has a greater impact on adrenaline responses in non-users than in habitual users, although habitual caffeine users, in general, tend to have a greater adrenaline response to exercise than non-users, even when just a placebo is consumed [24]; thus, caffeine intake may have both a short-term and long-term impact on adrenaline responses to exercise.

We found that both men and women tended to have higher glucose concentrations with caffeine ingestion compared with placebo intake (Fig. 2). In line with this, previous research has shown that there are no significant sex differences in caffeine responses, even between rest and exercise in the pharmacokinetics of caffeine [25]; however, caffeine has been shown to increase endurance time in women more than men [24], although this has been reported in absolute values where women ingested greater amounts of caffeine per body weight. As differences in oestrogen levels exist depending on the stage of menstruation [25], women in the present study may therefore have had different responses to caffeine depending on the timing of their menstrual cycle, which was not standardized in our study. Female reproductive hormones have been shown to influence both substrate metabolism during exercise [26] and caffeine metabolism [27]. Future studies on caffeine and exercise (or hypoglycaemia) should take into account menstrual phase and oral contraception use.

One major limitation of the present study was the fact that hypoglycaemia did not occur with any great frequency, even in the placebo visit. This makes it difficult to conclude that caffeine ingestion limits hypoglycaemia during exercise. In

fact, only one participant developed hypoglycaemia during exercise in our study (in the placebo visit) and had to stop exercising. The ingestion of 16 g of fast-active carbohydrates (Dex4©, AMG Medical Inc., Montreal, Canada) before exercise may explain the limited number of hypoglycaemic episodes during exercise in the placebo visit, at least until the very end of exercise (six of 13 participants developed hypoglycaemia in the placebo visit vs. two of 13 in the caffeine visit). With the decreasing trend in glucose concentrations during exercise, it is likely that more participants would have reached hypoglycaemia if endurance exercise continued for longer than 45 min. A second limitation worth noting is that only a small number of non-habitual caffeine users were examined in the present study (n=3) and it is generally believed that non-caffeine users have a greater adrenaline response to caffeine intake and therefore may derive more protection against hypoglycaemia. Indeed, in the three non-users, glucose levels were all higher after caffeine than after placebo intake. Further studies are needed, however, to determine the acute and sustained impact that caffeine consumption has on glucose counter-regulation during exercise in Type 1 diabetes.

In conclusion, the present study shows that acute caffeine ingestion modestly attenuated the drop in glycaemia during afternoon steady-state aerobic exercise in people with Type 1 diabetes. Importantly, however, caffeine ingestion promoted an elevation in blood glucose concentration at bedtime, but paradoxically was associated with lower glucose concentrations in the early morning hours the next day, which may increase post-exercise late-onset hypoglycaemia. Necessary precautions should be taken in the evening hours after caffeine ingestion to ensure safety in active individuals with Type 1 diabetes. A bedtime snack with carbohydrates and protein (i.e. dairy or yogurt-based snack) and more frequent monitoring and/or reduced basal rates or long-acting insulin in the evening hours may be needed after caffeine intake.

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Competing interests

None declared.

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References

1 Chimen M, Kennedy A, Nirantharakumar K, Pang TT, Andrews R, Narendran P. What are the health benefits of physical activity in type 1 diabetes mellitus? A literature review. *Diabetologia* 2012; **55**: 542–551.
2 Riddell M, Perkins BA. Type 1 diabetes and vigorous exercise: Applications of exercise physiology to patient management. *Can J Diabetes* 2006; **30**: 63–71.
3 Brazeau AS, Rabasa-Lhoret R, Strychar I, Mircescu H. Barriers to physical activity among patients with type 1 diabetes. *Diabetes Care* 2008; **31**: 2108–2109.
4 Galassetti P, Riddell MC. Exercise and type 1 diabetes (T1DM). *Compr Physiol* 2013; **3**: 1309–1336.
5 Rabasa-Lhoret R, Bourque J, Ducros F, Chiasson JL. Guidelines for premeal insulin dose reduction for postprandial exercise of different intensities and durations in type 1 diabetic subjects treated intensively with a basal-bolus insulin regimen (ultralente-lispro). *Diabetes Care* 2001; **24**: 625–630.
6 Spriet LL. Exercise and sport performance with low doses of caffeine. *Sports Med* 2014; **44**(S2): 175–184.
7 Zaharieva D, Riddell M. Caffeine and glucose homeostasis during rest and exercise in diabetes mellitus. *Appl Physiol Nutr Metab* 2013; **38**: 813–822.
8 Greer F, Friars D, Graham TE. Comparison of caffeine and theophylline ingestion: exercise metabolism and endurance. *J Appl Physiol* 2000; **89**: 1837–1844.
9 Richardson T, Ryder J, Thomas P, Kerr D. Influence of caffeine on frequency of hypoglycemia detected by continuous interstitial glucose monitoring system in patients with long-standing type 1 diabetes. *Diabetes Care* 2005; **28**: 1316–1320.
10 Watson JM, Sherwin RS, Deary IJ, Scott L, Kerr D. Dissociation of augmented physiological, hormonal and cognitive responses to hypoglycaemia with sustained caffeine use. *Clin Sci* 2003; **104**: 447–454.
11 Gallen I, Chitrabhanu B, Lumb A, Carr J. Caffeine supplementation reduces exercise induced decline in blood glucose and subseuqent hypoglycemia in adults with type 1 diabetes (T1DM) treated with multiple daily injections (MDI). *American Diabetes Association: 70th Scientific Sessions* 2010; Abstract no. 1184-P.
12 Zeman F, Ney D. Nutritional care in diabetes mellitus. Applications in medical nutrition therapy. Columbus, OH: Merrill, 1996, 225–228.
13 Riddell M, Burr J. Evidence-based risk assessment and recommendations for physical activity clearance: Diabetes mellitus and related comorbidities. *Appl Physiol Nutr Metab* 2011; **36**: S154–189.
14 Howley ET, Bassett DR, Welch HG. Criteria for maximal oxygen uptake: Review and commentary. *Med Sci Sports Exerc* 1995; **27**: 1292–1301.
15 Gledhill N, Cox D, Jamnik R. Endurance athletes' stroke volume does not plateau: Major advantage is diastolic function. *Med Sci Sports Exerc* 1994; **26**: 1116.
16 Péronnet F, Massicotte D. Table of nonprotein respiratory quotient: an update. *Can J Sport Sci* 1991; **16**: 23–29.
17 Seaquist ER, Anderson J, Childs B, Cryer P, Dagogo-Jack S, Fish L, et al. Hypoglycemia and diabetes: A report of a workgroup of the American Diabetes Association and the Endocrine Society. *Diabetes Care* 2013; **36**: 1384–1395.
18 Richardson T, Rozkovec A, Thomas P, Ryder J, Meckes C, Kerr D. Influence of caffeine on heart rate variability in patients with long-standing type 1 diabetes. *Diabetes Care* 2004; **27**: 1127–1131.
19 Costill DL, Dalsky GP, Fink WJ. Effects of caffeine ingestion on metabolism and exercise performance. *Med Sci Sports* 1978; **10**: 155–158.

20 Campbell MD, Walker M, Trenell MI, Stevenson EJ, Turner D, Bracken RM, et al. A low-glycemic index meal and bedtime snack prevents postprandial hyperglycemia and associated rises in inflammatory markers, providing protection from early but not late nocturnal hypoglycemia following evening exercise in type 1 diabetes. *Diabetes Care* 2014; **37**: 1845–1853.
21 Campbell MD, Walker M, Trenell MI, Jakovljevic DG, Stevenson EJ, Bracken RM, et al. Large pre- and postexercise rapid-acting insulin reductions preserve glycemia and prevent early- but not late-onset hypoglycemia in patients with type 1 diabetes. *Diabetes Care* 2013; **36**: 2217–2224.
22 Graham TE, Rush JWE, Vansoeren MH. Caffeine and exercise - metabolism and performance. *Can J Appl Physiol-Rev* 2001; **19**: 111–138.
23 Cornelis MC, El-Sohemy A, Kabagambe EK, Campos H. Coffee, CYP1A2 genotype, and risk of myocardial infarction. *J Am Med Assoc* 2006; **295**: 1135–1141.
24 Van Soeren MH, Sathasivam P, Spriet LL, Graham TE. Caffeine metabolism and epinephrine responses during exercise in users and nonusers. *J Appl Physiol* 1993; **75**: 805–812.
25 Graham TE, McLean C. Gender differences in the metabolic responses to caffeine. Gender Differences in Metabolism: Practical and Nutritional Implications. Boca Raton, FL: CRC Press, 1999: 301–327.
26 Oosthuyse T, Bosch AN. The effect of menstrual cycle on exercise metabolism: implications for exercise performance in eumenorrhoeic women. *Sports Med* 2010; **40**: 207–227.
27 Balogh A, Klinger G, Henschel L, Borner A, Vollanth R, Kuhnz W. Influence of ethinylestradiol-containing combination oral contraceptives with gestodene or levonorgestrel on caffeine elimination. *Eur J Clin Pharmacol* 1995; **48**: 161–166.

Supporting Information

Additional Supporting Information may be found in the online version of this article:

Figure S1. Pan-view of individual (delta) changes in whole-blood glucose concentrations (mmol/l) during exercise. Both placebo (triangles) and caffeine visit (circles) are shown as measured by self-monitoring of blood glucose. Letters represent individual participants and dashed lines represent mean values (***n***=13).