The Effect of Different Dosages of Caffeine on Endurance Performance Time

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The effect of different dosages of caffeine (0 – 5 – 9 – 13 mg · kg body weight^{-1}) on endurance performance was examined. Nine well-trained cyclists participated in this study (VO_2max 65.1 ± 2.6 ml · kg^{-1} · min^{-1}). Caffeine capsules were administered in random order and double-blind. One hour after capsule ingestion, subjects cycled until exhaustion at 80 % Wmax on an electromagnetically braked cycle ergometer. Blood samples were taken before, during and after the exercise test. Before and after the test a urine sample was obtained. A significant increase in endurance performance was found for all caffeine tests compared to placebo (endurance time 47 ± 13, 58 ± 11, 59 ± 12 and 58 ± 12 min for 0, 5, 9 and 13 mg · kg^{-1} body weight, respectively). No differences were found in endurance performance between the three caffeine dosages which indicates that no dose-response relation of caffeine and endurance performance was found. An increased free fatty acid and glycerol concentration was found after caffeine consumption compared with placebo. The mean urinary caffeine concentrations after exercise were 4.8 ± 1.8, 8.9 ± 5.2 and 14.9 ± 6.9 µg · ml^{-1} urine for 5, 9 and 13 mg of caffeine · kg^{-1} body weight. Only the lowest dose of caffeine resulted in urine caffeine concentrations below the doping limit of the International Olympic Committee of 12 µg · ml^{-1} urine in all individuals. It is concluded that caffeine is an ergogenic aid that stimulates endurance performance. A dose-response relation between caffeine and endurance time was not found for the dose-range investigated. The stimulating effect of caffeine was already apparent at the lowest dose of caffeine given (5 mg · kg^{-1}). At this dose urinary caffeine concentration remained below the doping limit in all subjects.

Key words: Caffeine, cycling exercise, doping, dose-response, endurance performance, urinary caffeine concentration

Introduction

The importance of the amount of glycogen stored in muscle cells for the endurance performance of athletes is well accepted (4,7,9). Increased fat metabolism during exercise enables subjects to perform longer, since glycogenolysis is decreased and glycogen is spared thereby delaying the moment of complete glycogen depletion and fatigue (3,4).

It has been suggested that caffeine is an ergogenic aid that increases lipolysis and fat oxidation and reduces glycogen breakdown. Because of its supposed ergogenic properties caffeine is on the doping list of the International Olympic Committee. However, the data on the metabolic and ergogenic effects of caffeine are equivocal. Costill et al. (8), Essig et al. (13) and Ivy et al. (17) demonstrated that caffeine ingestion resulted in a reduction of the exercise-induced breakdown of glycogen and enhanced use of blood borne free fatty acids (FFA) and/or muscle triglycerides. Other researchers, however, were unable to confirm the metabolic effects of caffeine (6,20,24,25).

The effects of caffeine on exercise performance are also controversial. Some studies found a stimulating effect of caffeine (8,17), others found no effect (20,24,25). Recent studies by Graham, Spriet and coworkers (13,26) showed a large increase of endurance performance after ingestion of a high dose of caffeine (9 mg · kg^{-1} body weight). Although the mean caffeine concentration in urine after exercise was below the doping limit of the International Olympic Committee (IOC), some urinary caffeine concentrations equalled or exceeded the doping limit of 12 µg · mol^{-1} urine.

In order to investigate whether the inconsistent findings described above were due to differences in the dosage of caffeine administered, the effect of three different dosages of caffeine on endurance performance and metabolic parameters was studied. In addition, urinary caffeine concentrations were measured after exercise, in order to establish the relationship between the amount of caffeine ingestion and urinary caffeine concentration after exercise more clearly. The urinary caffeine concentrations found in this study are discussed in relation to the doping limit of the IOC.

Int. J. Sports Med. 16 (1995) 225–230

Methods

Subjects

Nine healthy well-trained subjects were studied. Physical characteristics are given in Table 1. The experimental procedures and potential risks of the study were explained to each subject both verbally and in writing. All subjects gave informed consent. The experiment was approved by the Ethics Committee of the University of Limburg.

Table 1 Physical characteristics of the subjects participating in this study (n = 9).

Physical characteristics	mean ± SD
Height (cm)	181 ± 4
Weight (kg)	72.0 ± 4.0
Fat (%)	8.8 ± 2.1
Age (years)	22.1 ± 2.8
$\dot{V}O_2max$ ($l \cdot min^{-1}$)	4.7 ± 0.3
$\dot{V}O_2max$ ($ml \cdot min^{-1} \cdot kg^{-1}$)	65.1 ± 2.6
Wmax (W)	377 ± 27
80 % Wmax (W)	296 ± 22
HRmax (beats · min⁻¹)	189 ± 10
Wmax · BW^{-1} ($W \cdot kg^{-1}$)	5.24 ± 0.3

Pre-experimental protocol

Each subject visited the laboratory before the start of the actual study and performed an incremental $\dot{V}O_2max$ test on an electromagnetically braked cycle ergometer (Lode Excalibur, Groningen, The Netherlands) to determine maximal power output (Wmax) and maximal oxygen uptake ($\dot{V}O_2max$) (21). The mean $\dot{V}O_2max$ for the group and mean Wmax are shown in Table 1.

Instructions to the subjects

The subjects were instructed to maintain their normal training program during the study and were asked to incorporate the experimental test into their training program as a hard work out. The subjects were asked to refrain from training 24 h before the test. A diary was kept to record food consumption and activities during the three days before the first test. The subjects were asked to eat and train before the following tests as they had done before the first test to minimize variation in glycogen concentration in muscle and liver cells. An almost similar procedure has been shown to result in similar pretest glycogen concentrations (23). During these three pre-testing days subjects were asked to refrain from caffeine consumption (coffee/tea/chocolate(milk)/cola), as was done in the studies of Graham et al. and Spriet and colleagues (14,27), to make sure that no caffeine was present in the body before the exercise test. All subjects were caffeine users in varying gradations. Five subjects consumed between 100–250 mg caffeine per day and four subjects consumed above 250 mg caffeine per day.

Experimental protocol

Subjects performed the four endurance tests over four successive weeks. Day of the week and time of day were kept constant in each individual. The tests differed only in the amount of caffeine ingested before exercise. Besides a placebo that contained no caffeine (CAF) (0 mg CAF · kg body weight) the doses of caffeine tested were 5–9–13 mg CAF · kg body $weight^{-1}$ (CAF · kg BW^{-1}). The capsules were prepared and assigned by a chemist. The capsules were administered double-blindly and in random order.

Subjects arrived at 9.00 h or 13.00 h at the laboratory. Subjects who were tested in the morning had fasted, those who cycled in the afternoon were asked to fast from 11.00 h on. The subjects produced a urine sample immediately after arrival. A catheter was placed in an antecubital vein. One hour before exercise (t = – 60 min) a blood sample was taken and a capsule was ingested with water. After capsule ingestion subjects rested for an hour.

Before starting the exercise a blood sample was obtained (t = 0 min). Subjects cycled at 80 % Wmax until exhaustion. The test was ended when pedalling frequency was lower than 50 rpm. During the first 15 min of exercise a continuous sampling of expired gas was carried out and blood was sampled at 5, 10 and 15 minutes. After 15 min blood and gas samples were obtained every 10 min until exhaustion. Fifteen minutes after finishing the cycling exercise (recovery) another blood sample was obtained and urine was collected.

Analyses

Expired gas samples were analyzed every 20 sec for fractions of O_2 and CO_2 with an on-line system (Sensormedics 2900, Sensormedics Cooperation, USA). The blood samples were mixed with EDTA to prevent clotting and were centrifuged immediately. Plasma was frozen in liquid N_2 and stored at – 20 °C until further analysis. The urine samples were also stored at – 20 °C.

The [FFA] was determined with an *in vitro* enzymatic method for quantification of the [FFA] in plasma (Wako NEFAC kit; Wako Chemicals, Germany). The analyses were carried out with a Cobas-Bio semi-automated centrifugal spectrophotometer. This apparatus was also used for quantification of the glycerol concentration by standard enzymatic method. Caffeine concentrations in urine were determined using an HPLC method (18).

Data analysis

Data are presented as mean ± standard deviation (SD). Data of all subjects were available between – 60 and 35 min and these were used for statistical analysis. Data of the moment of exhaustion (EXH) and during recovery (REC) were also analyzed.

An analysis of variance (ANOVA) with repeated measurements was used for statistical analysis of the response differences between four dosages of caffeine. When significant differences were found ($p < 0.05$) the Scheffe F-test was used to test for differences between pairs of dosages. Pearson correlation coefficient (r) was calculated for relation between the dosage of caffeine and post-exercise urinary caffeine concentration.

Results

Time to exhaustion differed significantly among the caffeine and placebo tests. The endurance performance was signifi-

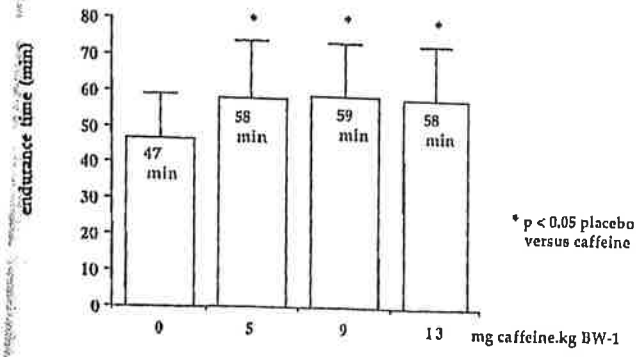

Fig. 1 Endurance performances with different caffeine concentrations. The cycling time (min) is averaged for 9 subjects. The caffeine performances were significantly better than the placebo performance. No significant differences among the caffeine tests were found.

Table 2 The individual endurance time data for the four different conditions in minutes and seconds. Mean and standard deviations of the four tests of the nine subjects are presented, too.

Subjects	0 mg	5 mg	9 mg	13 mg
1	36.05	42.47	51.50	37.55
2	52.47	85.15	65.00	59.30
3	56.55	63.20	73.10	79.12
4	45.20	52.10	64.40	58.33
5	35.25	66.20	57.45	70.54
6	66.38	73.25	76.49	69.47
7	40.57	44.50	40.55	46.48
8	57.15	57.17	66.47	66.35
9	28.34	35.05	33.17	36.20
mean	47	58	59	58
SD	13	11	12	12

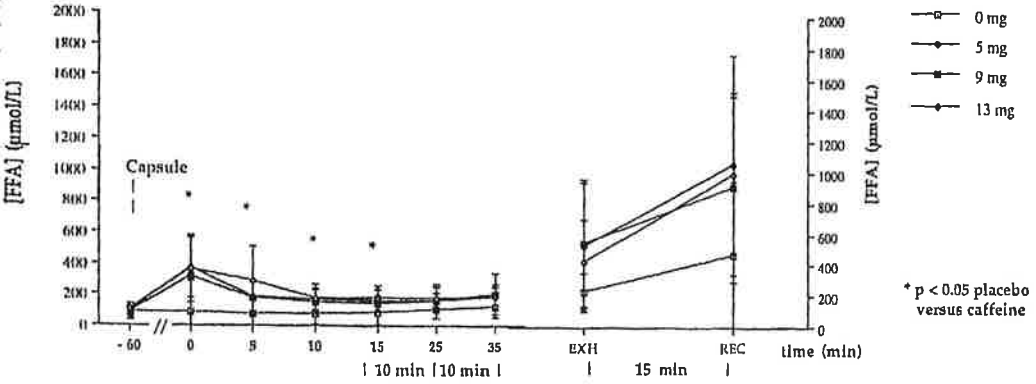

Fig. 2 Free fatty acid (FFA) concentrations during the 4 exercise tests. EXH: exhaustion REC: recovery

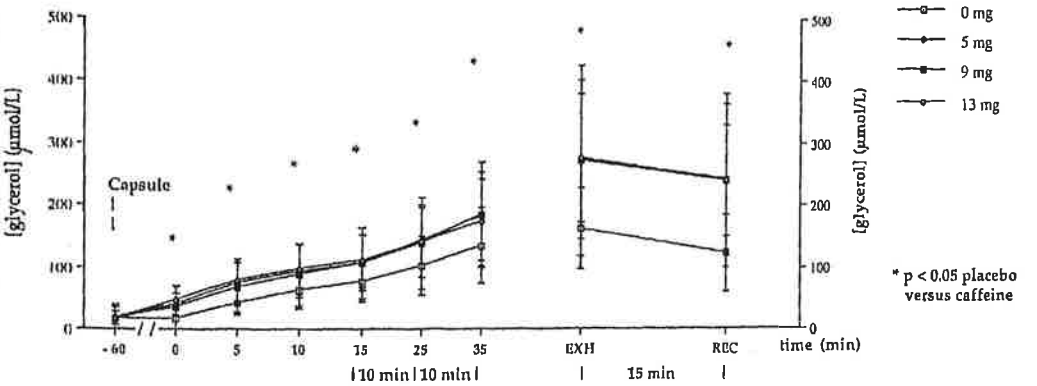

Fig. 3 Glycerol concentrations during the 4 exercise tests. EXH: exhaustion REC: recovery

cantly increased (average 27%) when caffeine was ingested compared to the placebo test (Fig. 1). Among the caffeine tests no significant differences in cycling time were found. Individual endurance performances are shown in Table 2.

Sixty minutes after caffeine ingestion (t = 0 min) FFA concentrations were significantly different from placebo (Fig. 2). During the first 15 min of exercise significantly higher FFA concentrations were found in all caffeine tests compared to placebo. No differences in FFA concentrations were found between the caffeine dosages. Similar to the FFA concentrations, the glycerol concentrations were significantly higher 60 min after caffeine ingestion (Fig. 3) compared to placebo. Unlike the FFA concentrations, the glycerol concentrations remained significantly higher after caffeine ingestion during the whole test and after the exercise test. The similarity of glycerol concentrations after caffeine intake is shown in Fig. 3. [The concentrations for the three caffeine concentrations are almost the same and therefore only one line is seen.]

The caffeine concentrations in urine following exercise showed a significant linear relationship with the amount of caffeine ingested before exercise ($r = 0.65$) ($p < 0.001$). The mean urinary caffeine concentration obtained after exercise was 4.8 ± 1.8, 8.9 ± 5.2 and $14.9 \pm 6.9\ \mu g \cdot ml^{-1}$ for 5, 9 and 13 mg CAF · kg BW^{-1}. The individual urinary caffeine concentrations after exercise are shown in Fig. 4 for the different caffeine dosages.

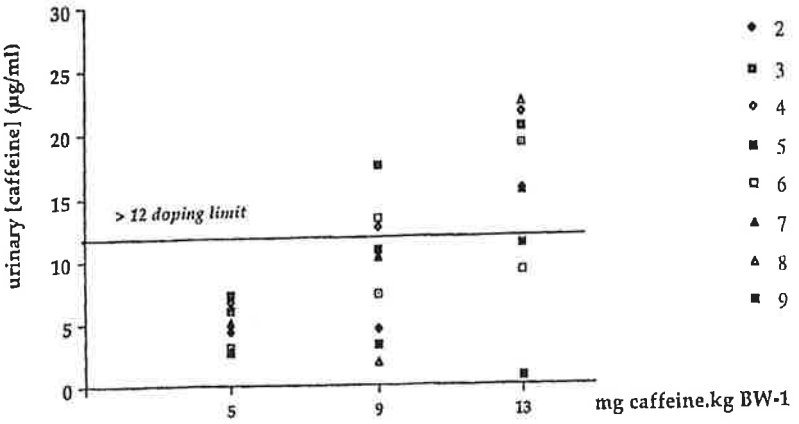

Fig. 4 Individual caffeine concentrations in urine obtained 15 minutes after finishing exercise. The doping level of the IOC is shown at 12 µg · ml^{-1} urine. Mean urinary caffeine concentrations obtained after exercise were 4.8 ± 1.8, 8.9 ± 5.2 and 14.9 ± 6.9 µg · ml^{-1} for 5, 9 and 13 mg · kg^{-1} caffeine.

Discussion

In the past decades the effect of caffeine on exercise performance has been examined in many studies. The studies showed equivocal results with respect to the proposed ergogenic effect of caffeine. Costill (8), Ivy (17) and coworkers found a stimulating effect of caffeine on endurance performance. However, other investigators like Knapik et al. (20), Perkins and Williams (24) and Powers and coworkers (25) did not find an ergogenic effect of caffeine.

Dose-response

Recent findings of Graham and Spriet (14,27) have again opened the discussion about the stimulating effect of caffeine during endurance performance. In their studies an endurance exercise test was performed at 80–85 % VO_2max after a high caffeine dose (9 mg · kg BW^{-1}) or placebo ingestion. For all subjects an increase in endurance performance was found, averaging 44 and 51 % for running and cycling, respectively. The subjects were highly trained or recreational athletes, men and women, caffeine users and non-users. The positive effect found may have been a consequence of the caffeine dose given before the exercise (14).

The effect of the caffeine dosage on endurance performance was examined in the present study. Some earlier studies investigating a dose-response relation for caffeine and endurance performance failed to show a relation (20,24). However, many researchers still suggest that a dose-response relation does exist (15,19).

In this study a wider range of caffeine dosages was used (0–5–9–13 mg CAF · kg BW^{-1}) to examine whether the 13 mg · kg BW^{-1} dose was more effective than the already high dose of 9 mg CAF · kg BW^{-1} dosage. Some professional cyclists use these large amounts of caffeine in competition (pers. comm.).

We found a stimulating effect of caffeine on cycling time for all three caffeine dosages (an average increase in performance time of 27 %). No differences were found between the caffeine tests; all caffeine dosages tested were equally effective with respect to endurance performance. The relative increase of 27 % in performance time is the same as found by Spriet et al. (27) for cycling exercise with a similar protocol. The differences in absolute endurance time found in this study and the study of Spriet et al. (27) (placebo 47 ± 13 min versus 76 ± 5 and for caffeine 58 ± 12 versus 96 ± 9, respectively), might be a consequence of the methods used to determine the workload during the test. In our study the subjects cycled at 80 % Wmax. Spriet et al. (27) determined Wmax and 30 minutes later subjects cycled for 10–15 min at a power output to elicit ≈ 80 % VO_2max. Because a levelling-off of the oxygen uptake at maximal work load is known, 80 % VO_2max might result in a lower work load than 80 % Wmax. It is possible that the mean workload differed considerably in these studies as a consequence of the methods used. In this study subjects cycled on average 296 W ± 22. This workload is high for an endurance performance. The mean work load of Spriet and coworkers (27) was not reported. A lower power output might explain the longer absolute endurance performance times as was shown by Spriet and colleagues (27).

The increase in performance time found in this study was lower than found by Graham and Spriet (14) for running (45 %) and cycling exercise (≈ 31 %, without subject 5 who showed an increase of 156 %). The results in this study are in agreement with the findings of Cadarette and coworkers (5). They investigated a dose-response relation (0–2.2–4.4–8.8 mg CAF · kg BW^{-1}) during running at 80 % VO_2max. The researchers concluded that no dose-response relation exists, although a significant difference between placebo and all caffeine trials was found. Studies carried out with lower amounts of caffeine (< 4 mg · kg BW^{-1}) showed conflicting results on endurance performance. Positive effects of caffeine were found by Wiles (29) using 150 to 200 mg caffeine, and Cadarette (5) using 200 to 300 mg caffeine, while Titlow (28) found no caffeine effect on submaximal exercise when 200 mg caffeine was ingested.

Mechanism of action of caffeine

The increase in endurance performance after ingestion of caffeine is thought to be caused by an enhanced fat oxidation (3, 8, 10, 11, 14, 15). An increase in fat oxidation during exercise will delay glycogen depletion and fatigue. The glycogen-sparing is

suggested to be especially effective in the first 15 min of exercise, because glycogenolysis is normally very high in this period (11). A simultaneous down-regulation of the CHO-metabolism (2) could result in glycogen-sparing which was also shown by Spriet et al. (27). In the present study FFA and glycerol concentrations in blood were increased after caffeine ingestion which is in agreement with previous findings in other caffeine studies (8,13,27). An increased lipolysis is implicated in the mechanism of action. In this study no dose-response relationship for FFA and glycerol was found.

The lipolysis might be increased via a direct or indirect caffeine effect. The cellular mechanisms of caffeine action were recently reviewed by Dodd et al. (12). The most probable cellular mechanism of caffeine is inhibition of adenosine receptors, thereby reducing the inhibition of lipolysis by adenosine. Lindinger et al. (22) suggested that the ergogenic effect of caffeine was caused by attenuation in plasma $[K^+]$ during exercise. Maintenance of membrane potential by Na-K pump activity may contribute to the stimulating effect of caffeine.

Ingestion of caffeine in competition

The results of the present study suggest that caffeine consumption before an endurance exercise positively influences performance. The maximal stimulating effect of caffeine was already found with the lowest dosage of caffeine (5 mg CAF · kg BW^{-1}). The urinary caffeine concentration was below the doping limit of 12 $\mu g \cdot ml^{-1}$ urine of the IOC for all subjects for the lowest dose of caffeine tested. This study showed that higher dosages of caffeine did not result in a better endurance performance, but did result in high urinary caffeine concentrations. The urinary caffeine concentrations were too high with respect to the doping limit for some individuals with the 9 mg CAF · kg BW^{-1} and for most of the subjects at the highest dosage given (see Fig. 4). There were large interindividual variations in the amount of caffeine in urine after exercise between the different caffeine dosages. Because caffeine distributes in the tissues in approximate proportion of the water content, the physiological response is influenced by hydration status and exercise duration (sweat production). This means that body composition (fat content), hydration state and caffeine dose ingested (10,15,26) are important factors with respect to the effect of caffeine. The amount of dehydration among the subjects may have differed during the exercise test. Caffeine further stimulates diuresis resulting in increased urine production, which could result in decreased caffeine concentration in urine. Another important aspect is the different endurance times resulting in different moments of urine sampling after exercise. Within 15 minutes after finishing the test urine was sampled. This might affect the amount of caffeine measured in urine. The acceptability of using urinary caffeine concentrations for competition screening has been questioned because of the variation in concentration measured after exercise (10). The doping limit of the IOC could be reached by ingesting 8 cups of coffee (100 $mg \cdot cup^{-1}$), indicating that athletes have to refrain from caffeine consumption before competition, because the ergogenic drug could otherwise be measured in a urine sample.

Caffeine consumption not only affects substrate metabolism, it is also known to effect mood and feelings of perceived exertion. In this study too, subjects reported that the exercise tests performed after caffeine consumption were easier to carry out than after placebo consumption.

Side effects of caffeine intake

The known side effects of caffeine like dizziness, headache, tremor, hunger sensations, insomnia and diuresis (4,17,20) were reported during and after the test, especially for the 9 and 13 mg CAF · kg BW^{-1} dosages, but disappeared within a few hours.

In conclusion, an ergogenic effect of caffeine is found for all caffeine dosages examined. The cycling time increased on average 27 % for all caffeine dosages. In the dose range investigated no dose-response relation was found. A maximal beneficial effect of caffeine was already found with 5 mg CAF · kg BW^{-1}. At this dosage all subjects had urinary caffeine concentrations below the doping limit of the IOC.

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