European Journal of Clinical Nutrition (2009) 63, 57–64
 0954-3007/09 $32.00
www.nature.com/ejcn

ORIGINAL ARTICLE

The effect of caffeine, green tea and tyrosine on thermogenesis and energy intake

A Belza, S Toubro and A Astrup

Department of Human Nutrition, Faculty of Life Sciences, University of Copenhagen, Copenhagen, Denmark

Objectives: To investigate the effect of three different food ingredients tyrosine, green tea extract (GTE) and caffeine on resting metabolic rate and haemodynamics, and on *ad libitum* energy intake (EI) and appetite.
Methods: Twelve healthy, normal weight men (age: 23.7 ± 2.6 years, mean ± s.d.) participated in a four-way crossover, randomized, placebo-controlled, double-blind study. Treatments were administered as tablets of 500 mg GTE, 400 mg tyrosine, 50 mg caffeine, or placebo, and were separated by >3-day washout. The acute thermogenic response was measured in a ventilated hood system for 4 h following ingestion. Blood pressure, heart rate (HR), and subjective appetite sensations were assessed hourly and *ad libitum* EI 4 h post-dose.
Results: Caffeine induced a thermogenic response of 6% above baseline value (72 ± 25 kJ per 4 h, mean ± s.e.) compared to placebo ($P<0.0001$). The thermogenic responses to GTE and tyrosine were not significantly different from placebo. Tyrosine tended to increase 4-h respiratory quotient by 1% compared to placebo (0.01 ± 0.005, $P=0.05$). *Ad libitum* EI was not significantly different between treatments but was reduced by 8% (−403 ± 183 kJ), 8% (−400 ± 335 kJ) and 3% (−151 ± 377 kJ) compared to placebo after intake of tyrosine, GTE and caffeine, respectively. No significant difference in haemodynamics was observed between treatments.
Conclusions: Only caffeine was thermogenic in the given dose and caused no haemodynamic side effects. The sample size was probably too small to detect any appetite suppressant properties of the treatments. Further investigations are required.
European Journal of Clinical Nutrition (2009) **63,** 57–64; doi:10.1038/sj.ejcn.1602901; published online 19 September 2007

Keywords: energy expenditure; energy intake; appetite; tyrosine; caffeine; green tea

Introduction

Long-term weight maintenance of a weight loss is often unsuccessful. There is a need for simple and safe methods to prevent weight gain and re-gain, for example by increasing energy expenditure and decreasing spontaneous energy intake (EI). Studies have shown that nutritional and food-related non-nutritional factors influence the adjustment of EI and energy expenditure as compounds can enhance diet-induced thermogenesis and suppress appetite. An increasing number of studies have examined the effects of well-known food ingredients on sympathetic nervous system (SNS) activity with an aim of preventing positive energy balance in humans (Kovacs and Mela, 2006). The activation of SNS has been shown to suppress hunger, enhance satiety and stimulate energy expenditure, in part by increasing fat oxidation (Astrup *et al*., 1991).

The thermogenic properties of green tea extract (GTE) have been the objective of several investigations. GTE contains catechins, which inhibit the enzyme catechol *O*-methyltransferase and thereby stimulate SNS. Catechol *O*-methyltransferase rapidly degrades noradrenalin (NA) in the synaptic cleft and lessens the adrenergic receptor stimulation of NA (Borchardt and Huber, 1975; Dulloo *et al*., 2000). Catechins from GTE, especially polyphenol catechin epigallocatechin gallate, have been associated with an increase in SNS activity, thermogenesis and fat oxidation in humans (Dulloo *et al*., 1999; Nagao *et al*., 2005), although with some inconsistencies (Kovacs *et al*., 2004).

The intracellular signal, producing increased lipolysis, heat production in skeletal muscle and putative satiety signals in the liver, is dependent on the production of cyclic adenosine monophosphate. The increased cyclic adenosine monophosphate response is short-lived, because cyclic adenosine

Correspondence: Dr A Belza, Department of Human Nutrition, The Centre for Advanced Food Studies, Faculty of Life Sciences, University of Copenhagen, Rolighedsvej 30, DK-1958 Frederiksberg C, Denmark.
E-mail: anbe@life.ku.dk.
Received 14 February 2007; revised 8 July 2007; accepted 20 July 2007; published online 19 September 2007

monophosphate is rapidly degraded by phosphodiesterase. The intracellular signal can be sustained for longer by the inhibition of phosphodiesterase by methylxanthines, which include caffeine. Caffeine also antagonizes the effect of adenosine in the presynaptic nerve terminals by reversing the adenosine-mediated inhibition of the release of neurotransmitters such as adrenaline and NA (Dulloo *et al*., 1994). GTE is rich in caffeine, which is also found in coffee, black tea and cacao. The acute effect of caffeine on thermogenesis ranges from 3 to 16% (Acheson *et al*., 1980; Hollands *et al*., 1981; Dulloo *et al*., 1989; Astrup *et al*., 1991, 1992; Arciero *et al*., 1995). Only one study has investigated the effect of caffeine on food intake (Tremblay *et al*., 1988). Epidemiological studies also suggest that caffeine might possess weight-reducing properties (Lopez-Garcia *et al*., 2006). However, there is solid evidence of caffeine being a potent amplifier of thermogenesis when given in conjunction with other SNS agonists such as ephedrine, nicotine, catechins or capsaicin from chillies (Dulloo *et al*., 1994; Yoshioka *et al*., 2001; Dulloo, 2002; Jessen *et al*., 2003, 2005; Zheng *et al*., 2004). The combination of catechins and caffeine in GTE is thought to both increase and prolong the effect of NA on SNS.

The amino acid tyrosine is an NA precursor and it may enhance NA synthesis and release in SNS (Rasmussen *et al*., 1983). However, it is believed that the rate of catecholamine synthesis is regulated by the rate-limiting enzyme tyrosine hydroxylase, which is strongly influenced by end-product inhibition via dopamine, adrenaline and NA. Tyrosine supplementation has decreased food intake in rats in a synergistic fashion when combined with other sympathomimetics (Hull and Maher, 1990, 1991, 1992). However, the effect of tyrosine supplementation in humans has not been investigated thoroughly.

The primary aim of the present study was to examine the individual effect of the compounds tyrosine, GTE and caffeine on thermogenesis, subjective appetite sensations, and *ad libitum* EI. The dose of the individual compounds was chosen from the existing literature as described above.

Methods and materials

Subjects

Twelve healthy and normal weight men (age: 23.7 ± 2.6 years, BMI: $22.4 \pm 1.8\,kg\,m^{-2}$, mean $\pm$ s.d.) participated in the study. They were weight-stable (± 3 kg in last 3 months), non-smoking, non-athletic and had no use of dietary supplements or frequent use of medication, and a low-to-moderate coffee intake. The subjects followed a normal Danish habitual diet, with rare use of hot spices, and avoided extreme intake of caffeine-containing beverages such coffee, tea, chocolate milk and some soft drinks. All subjects gave their written consent after having received verbal and written information about the study. The study was approved by The Municipal Ethical Committee of Copenhagen and Frederiksberg, and it was conducted in accordance with the Helsinki II Declaration.

Experimental design

The present study was designed as a four-way crossover, randomized, placebo-controlled, double-blind study. Each treatment was separated by >3-day washout period. All treatments were administered as tablets containing either 500 mg GTE (whereof 125 mg catechins), 400 mg tyrosine, 50 mg anhydrous caffeine or placebo. The relatively low dose of caffeine was chosen because of experience from our previous studies with caffeine, and to avoid any side effects (Astrup *et al*., 1990).

The placebo tablets contained microcrystalline cellulose and could not be distinguished from the bioactive treatment. The treatments were similarly encapsulated and differed only with regard to the content of the individual bioactive ingredient or placebo vehicle. The subjects were not allowed to change their dietary and beverage habits (including intake of coffee and tea), use of spices, level of physical activity, smoking habits and use of medication, throughout the study period.

Respiratory measurements

On each treatment day, the subjects arrived at the department at 0800 hours. After voiding (emptying bladder), height and body weight were measured to the nearest 0.05 kg on a decimal scale (Lindeltronic 8000, Copenhagen, Denmark) and height to the nearest 0.5 cm. The subjects rested for at least half an hour before undergoing assessments of resting metabolic rate (RMR) and respiratory quotient (RQ) by indirect calorimetry using a ventilated hood system (described in detail in Astrup *et al*., 1991). RMR was calculated using a formula assuming a fixed protein catabolism (Weir, 1949). The precision of the ventilated hood was validated by an alcohol burning test on a weekly basis; coefficient of variance (CV) was 1.5.

The respiratory measurements were of 4.5-h duration, from 0830 to 1300 hours. Between 0830 and 900 hours, a baseline measurement (25 min) was assessed. At around 0900 hours, the participants ingested one of the four treatment compounds together with 175 ml tap water and 25-min respiratory measurements were commenced. These were repeated eight times during the next 4 h (post-dose). Blood pressure (BP) and heart rate (HR) were assessed at 0830, 0930, 1030, 1130, 1230 and 1300 hours. The 175 ml tap water ingested together with the test compounds contained 21 mg calcium, primarily as $CaCO_3$. Although high doses of calcium seem to have an anti-obesity effect (Zemel, 2002), the present dose seems too small to have an effect on thermogenesis, substrate oxidation or food intake (Lorenzen *et al*., 2007).

The participants were instructed to fast, except for 0.5 l of water, from 2000 hours in the evening prior to the measurement. The subjects abstained from other than habitual medication, and from alcohol and energetic physical activity for 24 h before the respiratory measurements. To limit diurnal variation and inter- and intra-subject

variations, all measurements were carried out according to an identical schedule and at the same time of day.

Subjective appetite sensations
Visual analogue scales (VAS, described previously in Flint *et al*., 2000) were used to monitor each subject's appetite sensations before and after intake of the test compound. The scales contained questions about subjective sensations of hunger, satiety, prospective consumption, fullness and desire to eat something sweet, salty, rich in fat, or meat/fish. The subjects were instructed to complete VAS immediately before intake of the compound and at 1000, 1100, 1200 and 1300 hours.

Ad libitum *energy intake*
At completion of the respiratory measurements, the subjects were given an *ad libitum* brunch, 4 h after intake of one of the treatment compounds. The *ad libitum* meal was a 1329 g pasta salad brunch (610 kJ 100 g^{-1}, protein: 15 E%, carbohydrates: 55 E%, fat: 30%). The subjects were instructed to eat at a constant pace and to stop eating when they felt satiated. *Ad libitum* EI was assessed from the amount of the meal consumed.

Statistical analysis
All descriptive data are given in mean and standard deviation (s.d.). All results are given in mean and standard error (s.e.). The level of significance was set at <0.05. Statistical analyses were performed with SAS 8.2 (SAS Institute, Cary, NC, USA). All data were analysed as intention-to-treat and the last observation was carried forward. Prior to the statistical analysis, all data were tested for normality by Shapiro–Wilk W-test and variance homogeneity and data transformed if necessary. Differences between supplements were tested by analysis of mixed linear models procedure as repeated measurement adjusted for baseline level, and with or without adjusting for other confounders. *Post hoc* comparisons were made, with Turkey–Kramer adjustment of significance levels for the pair-wise comparison, using unpaired *t*-test when the analysis indicated significant treatment effect. Differences between baseline levels were tested by analysis of mixed linear models procedure.

Four-hour baseline-subtracted values of RMR, RQ and VAS scores were also calculated as an area under the curve (AUC), and difference between treatments tested by mixed linear models procedure.

Results

Body weight
There were no significant differences in mean body weight between treatments (GTE: 76.6±1.9 kg, tyrosine: 76.4±1.8 kg, caffeine: 76.6±1.9 kg, placebo: 76.3±1.8 kg, $P=0.7$).

Resting metabolic rate and respiratory quotient
There was no periodic effect in 4 h RMR or RQ, or any interaction between treatments and the previous treatment (carry-over effect). No significant difference between treatments was observed on baseline levels of RMR or RQ (Table 1). The caffeine treatment induced a thermogenic response of 72 (21:123) kJ per 4 h (mean, 95% CI) greater than placebo ($P<0.0001$) (Figure 1a). After placebo subtraction, the caffeine induced a RMR response of 6% above basal metabolic rate (BMR) baseline value (72±25 kJ per 4 h,

Table 1 The baseline level of respiratory measures, haemodynamic measures and subjective appetite ratings in 12 young men (mean±s.e.)[a]

	Green tea	*Tyrosine*	*Caffeine*	*Placebo*
Respiratory measures				
Thermogenesis, kJ day^{-1}	7651±157	7535±272	7266±272	7590±251
Respiratory quotient	0.87±0.02	0.86±0.02	0.85±0.02	0.86±0.01
Haemodynamic measures				
Systolic blood pressure, mm Hg	114±2	115±3	116±4	110±3
Diastolic blood pressure, mm Hg	61±2	61±2	62±2	61±2
Heart rate, bpm	53±2	53±3	53±3	53±3
Visual analogue scales				
Satiety, cm	3.7±0.4	3.4±0.4	3.2±0.3	3.7±0.3
Fullness, cm	2.8±0.4	3.2±0.6	2.4±0.3	2.8±0.3
Prospective consumption, cm	6.6±0.3	6.7±0.6	7.0±0.3	6.4±0.3
Hunger, cm	6.1±0.7	6.0±0.7	7.0±0.2	6.0±0.5
Salty, cm	4.5±0.3	4.6±0.7	4.0±0.5	4.6±0.5
Sweet, cm	3.4±0.4	3.9±0.6	3.1±0.4	3.4±0.4
Fat, cm	4.7±0.5	5.1±0.7	4.4±0.5	4.6±0.5
Meat/fish, cm	4.4±0.7	4.8±0.8	4.1±0.4	4.5±0.4

[a]Data were analysed by analysis of variance (mixed linear models). No significant difference was observed between treatments.

$P=0.01$), equivalent to $6\times72=432$ kJ on a 24-h basis (Figure 1b). No thermogenic effect of tyrosine or GTE supplementation was observed (Table 2). Tyrosine and GTE induced RMR responses 2% above BMR baseline (66 ± 71 kJ day^{-1}, $P=0.1$) and 0.4% (15 ± 65 kJ day^{-1}, $P=0.4$) compared to placebo, respectively.

When testing the results by repeated measurement analysis of variance, tyrosine tended to increase carbohydrate oxidation by inducing a 1% greater 4-h RQ response (0.01 ± 0.005, $P=0.05$) compared to placebo (Figure 2a). However, this result was not reproduced when testing the difference between treatments calculated as AUC (Figure 2b and Table 2). No significant difference was found between the other treatments.

Blood pressure and heart rate

HR and diastolic BP were in general greater after caffeine 4 h post-intake compared to the other treatments. However, no significant treatment effects were seen in any of the haemodynamics (Table 2). The change in HR and RMR after intake of caffeine correlated significantly ($r=0.9$, $P=0.003$). No other correlations were found between change in HR and RMR after intake of either tyrosine GTE or placebo. Even though treatment period and carry-over effect exerted a confounding effect ($P=0.02$ and $P=0.002$) on HR, no alteration of the present result was found when adjusted with or without the two confounders. No significant difference between treatments was observed on baseline levels of BP or HR (Table 1).

Ad libitum *energy intake*

Four hours after intake of GTE, tyrosine and caffeine *ad libitum* EI was decreased insignificantly by 8% (-400 ± 335 kJ, $P=0.5$), 8% (-403 ± 183 kJ, $P=0.3$) and 3% (-151 ± 377 kJ, $P=0.7$) respectively, compared to placebo (4859 ± 431 kJ).

Appetite sensations

VAS ratings of satiety, fullness, prospective consumption and hunger showed no significant difference between treatments. The desire to eat something salty was lower after intake of tyrosine compared to placebo ($P=0.04$). The desire to eat something sweet and something fat was greater 4 h post-intake of GTE compared to placebo ($P=0.008$ and $P=0.04$, respectively). The desire to eat something fat was significantly higher 4 h post-intake of GTE compared to placebo. No significant difference between treatments was observed on any of the baseline levels of the subjective appetite sensations (Table 1).

Discussion

The present dose of caffeine induced a 6% thermogenic response post-intake, which supports previous observations of the thermogenic properties of caffeine. However, the ingested 50 mg caffeine is of rather low dosage compared to previous studies where the acute dose has generally been between 100 and 400 mg caffeine, inducing a thermogenic response between 3 and 16% compared to the control (placebo or decaffeinated coffee) (Acheson *et al*., 1980; Hollands *et al*., 1981; Dulloo *et al*., 1989; Astrup *et al*., 1991; Arciero *et al*., 1995). Caffeine may possess both thermogenic and appetite-suppressant effects (Jessen *et al*., 2003, 2005), and together these effects on energy might produce weight loss in habitual users. This is supported by epidemiological studies suggesting that caffeine intake is inversely associated with prospective weight change. Lopez-

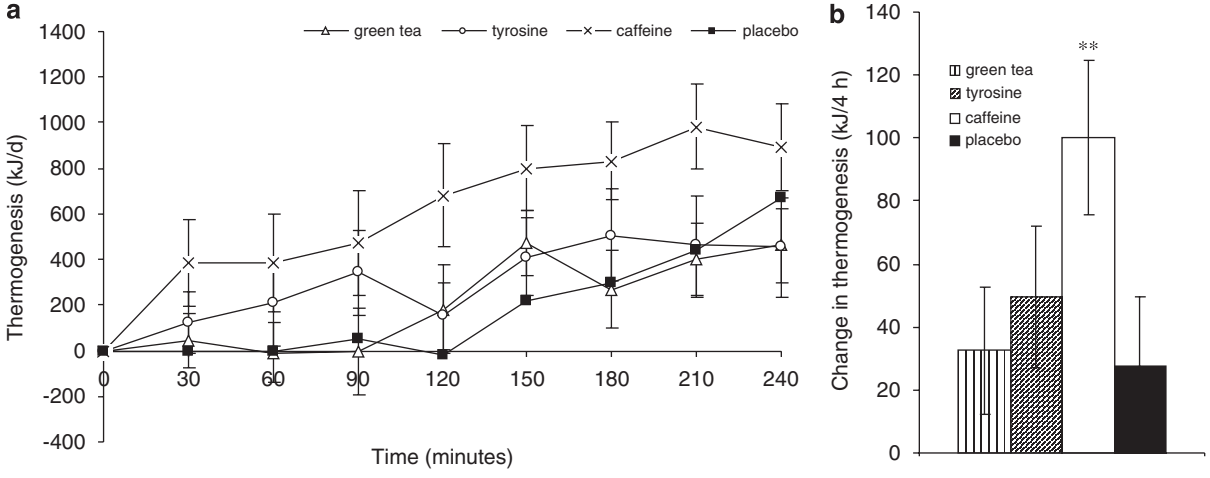

Figure 1 Baseline-subtracted 4 h thermogenic response (presented as (**a**) repeated measurement and (**b**) area under the curve (AUC)) in 12 young men after treatment with green tea, placebo, tyrosine or caffeine. Data are presented as mean$\pm$s.e. and were analysed as repeated measurements and dAUC by analysis of variance (mixed linear models). A significant difference was found between caffeine and placebo ($P<0.0001$).

Table 2 The level of baseline-subtracted 4 h post-intake change (4 h dAUC) in thermogenic response, respiratory quotient and haemodynamics in 12 young men (mean ± s.e.)[a]

	Green tea	*Tyrosine*	*Caffeine*	*Placebo*
Thermogenesis, kJ	33 ± 20	50 ± 22	100 ± 25[b]	28 ± 22
Respiratory quotient	0.01 ± 0.02	0.04 ± 0.03	−0.02 ± 0.02	−0.01 ± 0.03
Systolic blood pressure, mm Hg	0.8 ± 5.6	−8.6 ± 7.8	2.9 ± 7.8	9.0 ± 4.8
Diastolic blood pressure, mm Hg	−1.8 ± 5.7	−1.9 ± 2.7	1.9 ± 4.2	−5.9 ± 3.7
Heart rate, bpm	−376 ± 287	−50 ± 223	157 ± 294	−40 ± 355

Abbreviation: AUC, area under the curve.
[a]Data were analysed by analysis of variance (mixed linear models).
[b]The 4 h thermogenic response of caffeine was significantly different from placebo ($P < 0.0001$).

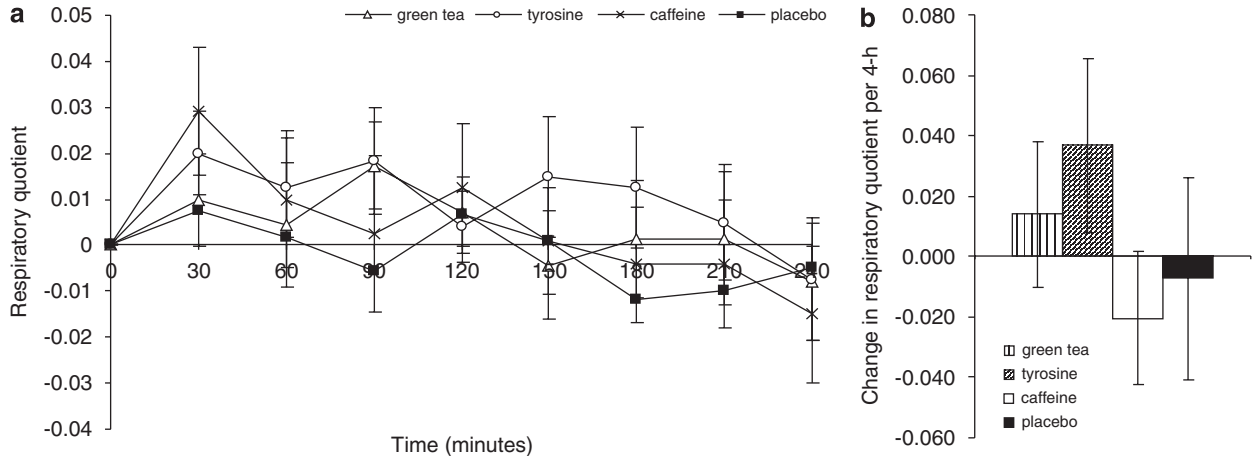

Figure 2 Baseline-subtracted 4 h change in respiratory quotient (presented as (**a**) repeated measurement and (**b**) area under the curve (AUC)) in 12 young men after treatment with green tea, placebo, tyrosine or caffeine. Data are presented as mean ± s.e. and were analysed as repeated measurements and dAUC by analysis of variance (mixed linear models).

Garcia *et al*. (2006) have found an association between weight regain and habitual coffee consumption, whereas other studies have not confirmed this finding (Astrup *et al*., 1992; Pasman *et al*., 1997). This may be influenced by the fact that natural sources of caffeine also contain widely varying mixtures of other xanthines and of other substances, such as tannins, which could affect energy balance (Magkos and Kavouras, 2005).

The rather high thermogenic response to 50 mg caffeine in the present study may partly be explained by the subjects' relatively low habitual intake of coffee. It has been speculated that long-term high intake of caffeine or other methylxanthines causes insensitivity to the effects of caffeine. High- and low-caffeine consumers have been shown to have different responses to caffeine, and high-caffeine consumers seem to be less stimulated by caffeine or other sympathomimetics such as green tea (Kovacs *et al*., 2004; Westerterp-Plantenga *et al*., 2005; Diepvens *et al*., 2007).

Caffeine is thought to increase SNS activity by stimulating the α- and β-adrenoreceptors, and to induce adenosine antagonism, which can lead to increased BP and HR (Astrup and Toubro, 1993; Dulloo, 1993). This side effect was not found in the present study but it has been observed in other studies with larger doses of supplemented caffeine (Astrup *et al*., 1990; Arciero *et al*., 1998; Noordzij *et al*., 2005). However, the evidence of the effect of caffeine on haemodynamics is inconsistent (Astrup *et al*., 1991, 1992; Toubro *et al*., 1993). Interestingly, the caffeine-induced increase in haemodynamics seems to be diminished by sustained caffeine-intake over time (James, 1994; Debrah *et al*., 1995). This suggests that subjects may adapt to a high habitual or increased caffeine intake.

The lack of thermogenic effect of GTE found in the present study supports other studies investigating the effect of catechins on energy expenditure (Kovacs *et al*., 2004; Diepvens *et al*., 2005; Klaus *et al*., 2005). However, most studies examining the effect of tea catechins have found that the agents were able to enhance thermogenesis even at smaller doses than those in present study (Dulloo *et al*., 1999, 2000; Berube-Parent *et al*., 2005; Westerterp-Plantenga *et al*., 2005), which suggests that tea catechins possess thermogenic properties. However, the effect of catechins seems dependent on a synergy with caffeine. The caffeine content in green tea does not seem to be the main contributor of the green tea-induced thermogenic response. Dulloo *et al*. (1999,

2000) observed that green tea could stimulate thermogenesis to a much greater extent than that which can be attributed to the caffeine content alone. It has been suggested that the bioactivity of green tea is very much dependent on the concentration of the different types of catechins. Especially epigallocatechin gallate has been shown to be the most pharmacologically active, and supplementation has resulted in enhanced SNS activity (though not consistently; Kovacs *et al*., 2004) and thermogenesis, and reduced fat accumulation in humans (Dulloo *et al*., 1999; Berube-Parent *et al*., 2005; Nagao *et al*., 2005) and rodents (Dulloo *et al*., 2000; Klaus *et al*., 2005; Wolfram *et al*., 2005). However, in the present study, the relative contribution of the specific catechin isomers was not known. This may explain the lack of thermogenic response.

Intake of 400 mg tyrosine did cause a 2%, but nonsignificant, thermogenic response in 4 h RMR. Tyrosine, given separately to rodents, seems to have only a weak effect on SNS activity. However, Hull and Maher (1991) have shown that tyrosine given in combination with other sympathomimetics, such as phenylpropnolamine, ephedrine and amphetamine, could significantly increase temperature in the brown adipose tissue in rodents. This suggests that tyrosine may enhance the effect of other SNS agonists synergistically. However, the thermogenic effect of tyrosine in humans is still unknown and should be investigated in future studies.

In a rodent study, tyrosine has been shown to increase SNS activity and to suppress food intake in a synergistic and dose-dependent manner when administered with the above sympathomimetics (Hull and Maher, 1990). These results support the present study in which tyrosine caused an 8% decrease in *ad libitum* EI, although this was not significant. In addition, the administration of GTE in the present study also caused an 8% but insignificant decrease in EI. Despite the insignificant effects on EI, it should be emphasized that from a quantitative standpoint, the effects of tyrosine and GTE on EI largely exceed the stimulating thermogenic effect of these compounds. This suggests that a suppression of EI produced by tyrosine and GTE may be more important than a possible thermogenic effect. This pattern has also been observed with capsaicin, the pungent principle in chili pepper, when it is added to meals (Yoshioka *et al*., 1999, 2001). However, the significantly greater desire to ingest more fat and sweet foods following GTE supplementation may, in real life, offset the potential gains from a decreased EI. Only few studies have investigated a possible anorectic effect of GTE and the evidence is contradicting in both rodent studies (Kao *et al*., 2000; Klaus *et al*., 2005) and human studies (Kovacs *et al*., 2004; Diepvens *et al*., 2005; Westerterp-Plantenga *et al*., 2005).

The present supplementation of caffeine did not reduce *ad libitum* EI, which contradicts the study of Tremblay *et al*. (1988), who found that a supplement of 300 mg caffeine decreased *ad libitum* EI by 22% in men but not in women. However, in the present study, only 50 mg caffeine was administered, which was probably too low a dose to induce an anorectic effect. In addition, the present results of *ad libitum* EI must be regarded with caution. The 4 h delay between the ingredient ingestion and the *ad libitum* meal may have been too long to permit an optimal assessment of the impact of the ingredients on spontaneous EI.

Neither caffeine nor GTE administration in the present study showed any effect on substrate oxidations. Human studies have shown that catechins and caffeine can increase fat oxidation (Acheson *et al*., 1980; Dulloo *et al*., 1999; Westerterp-Plantenga *et al*., 2005). However, there are inconsistencies in the evidence of changes in fat utilization caused by caffeine or catechins (Astrup *et al*., 1991; Arciero *et al*., 1995; Bracco *et al*., 1995; Kovacs *et al*., 2004). The present tyrosine supplementation tended to significantly increase RQ/4 h by 1% when tested as repeated measurements. However, this must be regarded with caution as the result was not reproduced when testing the difference as AUC. It is not possible to compare the present finding with results from other studies as the effect of tyrosine on substrate oxidation has not been tested previously. The increase in RQ also goes against the assumption that tyrosine supplementation, despite the rate-limiting effect of tyrosine hydroxylase, may cause increased SNS activity mediated by stimulus of NA on β-adrenergic receptors.

In conclusion, only caffeine in the given dose was thermogenic and without causing any haemodynamic or other side effects. Although the reductions in *ad libitum* EI were not significant, they support previous findings. The small sample size can have prevented the detection of any appetite suppressant properties of the treatments and further investigations are required.

Acknowledgements

We thank John Lind for his expert technical assistance and Jens Kondrup for critical reading of the manuscript. The study was supported by a grant from Science, Toxicology & Technology, San Francisco, CA, USA. The dietary supplements containing the ingredients examined in the present paper were manufactured by Alpine Health Products, Salt Lake City, UT, USA, and are not commercially available.

References

Acheson KJ, Zahorska-Markiewicz B, Pittet P, Anantharaman K, Jequier E (1980). Caffeine and coffee: their influence on metabolic rate and substrate utilization in normal weight and obese individuals. *Am J Clin Nutr* **33**, 989–997.

Arciero PJ, Gardner AW, Benowitz NL, Poehlman ET (1998). Relationship of blood pressure, heart rate and behavioral mood state to norepinephrine kinetics in younger and older men following caffeine ingestion. *Eur J Clin Nutr* **52**, 805–812.

Arciero PJ, Gardner AW, Calles-Escandon J, Benowitz NL, Poehlman ET (1995). Effects of caffeine ingestion on NE kinetics, fat oxidation, and energy expenditure in younger and older men. *Am J Physiol* **268**, E1192–E1198.

Astrup A, Toubro S (1993). Thermogenic, metabolic, and cardiovascular responses to ephedrine and caffeine in man. *Int J Obes Relat Metab Disord* **17** (Suppl 1), S41–S43.

Astrup A, Breum L, Toubro S, Hein P, Quaade F (1992). The effect and safety of an ephedrine/caffeine compound compared to ephedrine, caffeine and placebo in obese subjects on an energy restricted diet. A double blind trial. *Int J Obes Relat Metab Disord* **16**, 269–277.

Astrup A, Toubro S, Cannon S, Hein P, Madsen J (1991). Thermogenic synergism between ephedrine and caffeine in healthy volunteers: a double-blind, placebo-controlled study. *Metabolism* **40**, 323–329.

Astrup A, Toubro S, Cannon S, Hein P, Breum L, Madsen J (1990). Caffeine: a double-blind, placebo-controlled study of its thermogenic, metabolic, and cardiovascular effects in healthy volunteers. *Am J Clin Nutr* **51**, 759–767.

Berube-Parent S, Pelletier C, Dore J, Tremblay A (2005). Effects of encapsulated green tea and Guarana extracts containing a mixture of epigallocatechin-3-gallate and caffeine on 24 h energy expenditure and fat oxidation in men. *Br J Nutr* **94**, 432–436.

Borchardt RT, Huber JA (1975). Catechol *O*-methyltransferase. 5. Structure–activity relationships for inhibition by flavonoids. *J Med Chem* **18**, 120–122.

Bracco D, Ferrarra JM, Arnaud MJ, Jequier E, Schutz Y (1995). Effects of caffeine on energy metabolism, heart rate, and methylxanthine metabolism in lean and obese women. *Am J Physiol* **269**, E671–E678.

Debrah K, Haigh R, Sherwin R, Murphy J, Kerr D (1995). Effect of acute and chronic caffeine use on the cerebrovascular, cardiovascular and hormonal responses to orthostasis in healthy volunteers. *Clin Sci (Lond)* **89**, 475–480.

Diepvens K, Kovacs EM, Nijs IM, Vogels N, Westerterp-Plantenga MS (2005). Effect of green tea on resting energy expenditure and substrate oxidation during weight loss in overweight females. *Br J Nutr* **94**, 1026–1034.

Diepvens K, Westerterp KR, Westerterp-Plantenga MS (2007). Obesity and thermogenesis related to the consumption of caffeine, ephedrine, capsaicin and green tea. *Am J Physiol Regul Integr Comp Physiol* **292**, R77–R85.

Dulloo AG (1993). Ephedrine, xanthines and prostaglandin-inhibitors: actions and interactions in the stimulation of thermogenesis. *Int J Obes Relat Metab Disord* **17** (Suppl 1), S35–S40.

Dulloo AG (2002). Herbal simulation of ephedrine and caffeine in treatment of obesity. *Int J Obes Relat Metab Disord* **26**, 590–592.

Dulloo AG, Duret C, Rohrer D, Girardier L, Mensi N, Fathi M *et al.* (1999). Efficacy of a green tea extract rich in catechin polyphenols and caffeine in increasing 24-h energy expenditure and fat oxidation in humans. *Am J Clin Nutr* **70**, 1040–1045.

Dulloo AG, Geissler CA, Horton T, Collins A, Miller DS (1989). Normal caffeine consumption: influence on thermogenesis and daily energy expenditure in lean and postobese human volunteers. *Am J Clin Nutr* **49**, 44–50.

Dulloo AG, Seydoux J, Girardier L (1994). Paraxanthine (metabolite of caffeine) mimics caffeine's interaction with sympathetic control of thermogenesis. *Am J Physiol* **267**, E801–E804.

Dulloo AG, Seydoux J, Girardier L, Chantre P, Vandermander J (2000). Green tea and thermogenesis: interactions between catechin-polyphenols, caffeine and sympathetic activity. *Int J Obes Relat Metab Disord* **24**, 252–258.

Flint A, Raben A, Blundell JE, Astrup A (2000). Reproducibility, power and validity of visual analogue scales in assessment of appetite sensations in single test meal studies. *Int J Obes Relat Metab Disord* **24**, 38–48.

Hollands MA, Arch JR, Cawthorne MA (1981). A simple apparatus for comparative measurements of energy expenditure in human subjects: the thermic effect of caffeine. *Am J Clin Nutr* **34**, 2291–2294.

Hull KM, Maher TJ (1990). -tyrosine potentiates the anorexia induced by mixed-acting sympathomimetic drugs in hyperphagic rats. *J Pharmacol Exp Ther* **255**, 403–409.

Hull KM, Maher TJ (1991). -tyrosine fails to potentiate several peripheral actions of the sympathomimetics. *Pharmacol Biochem Behav* **39**, 755–759.

Hull KM, Maher TJ (1992). Effects of L-tyrosine on mixed-acting sympathomimetic-induced pressor actions. *Pharmacol Biochem Behav* **43**, 1047–1052.

James JE (1994). Chronic effects of habitual caffeine consumption on laboratory and ambulatory blood pressure levels. *J Cardiovasc Risk* **1**, 159–164.

Jessen A, Buemann B, Toubro S, Skovgaard IM, Astrup A (2005). The appetite-suppressant effect of nicotine is enhanced by caffeine. *Diabetes Obes Metab* **7**, 327–333.

Jessen AB, Toubro S, Astrup A (2003). Effect of chewing gum containing nicotine and caffeine on energy expenditure and substrate utilization in men. *Am J Clin Nutr* **77**, 1442–1447.

Kao YH, Hiipakka RA, Liao S (2000). Modulation of endocrine systems and food intake by green tea epigallocatechin gallate. *Endocrinology* **141**, 980–987.

Klaus S, Pultz S, Thone-Reineke C, Wolfram S (2005). Epigallocatechin gallate attenuates diet-induced obesity in mice by decreasing energy absorption and increasing fat oxidation. *Int J Obes (Lond)* **29**, 615–623.

Kovacs EM, Mela DJ (2006). Metabolically active functional food ingredients for weight control. *Obes Rev* **7**, 59–78.

Kovacs EM, Lejeune MP, Nijs I, Westerterp-Plantenga MS (2004). Effects of green tea on weight maintenance after body-weight loss. *Br J Nutr* **91**, 431–437.

Lopez-Garcia E, van Dam RM, Rajpathak S, Willett WC, Manson JE, Hu FB (2006). Changes in caffeine intake and long-term weight change in men and women. *Am J Clin Nutr* **83**, 674–680.

Lorenzen JK, Nielsen S, Holst JJ, Tetens I, Rehfeld JF, Astrup A (2007). Effect of dairy calcium or supplementary calcium intake on postprandial fat metabolism, appetite, and subsequent energy intake. *Am J Clin Nutr* **85**, 678–687.

Magkos F, Kavouras SA (2005). Caffeine use in sports, pharmacokinetics in man, and cellular mechanisms of action. *Crit Rev Food Sci Nutr* **45**, 535–562.

Nagao T, Komine Y, Soga S, Meguro S, Hase T, Tanaka Y *et al.* (2005). Ingestion of a tea rich in catechins leads to a reduction in body fat and malondialdehyde-modified LDL in men. *Am J Clin Nutr* **81**, 122–129.

Noordzij M, Uiterwaal CS, Arends LR, Kok FJ, Grobbee DE, Geleijnse JM (2005). Blood pressure response to chronic intake of coffee and caffeine: a meta-analysis of randomized controlled trials. *J Hypertens* **23**, 921–928.

Pasman WJ, Westerterp-Plantenga MS, Saris WH (1997). The effectiveness of long-term supplementation of carbohydrate, chromium, fibre and caffeine on weight maintenance. *Int J Obes Relat Metab Disord* **21**, 1143–1151.

Rasmussen DD, Ishizuka B, Quigley ME, Yen SS (1983). Effects of tyrosine and tryptophan ingestion on plasma catecholamine and 3, 4-dihydroxyphenylacetic acid concentrations. *J Clin Endocrinol Metab* **57**, 760–763.

Toubro S, Astrup AV, Breum L, Quaade F (1993). Safety and efficacy of long-term treatment with ephedrine, caffeine and an ephedrine/caffeine mixture. *Int J Obes Relat Metab Disord* **17** (Suppl 1), S69–S72.

Tremblay A, Masson E, Leduc S, Houde A, Després J-P (1988). Caffeine reduces spontaneous energy intake in men but not in women. *Nutr Res* **8**, 553–558.

Weir JB (1949). New methods for calculating metabolic rate with special reference to protein metabolism. *J Physiol* **109**, 1–9.

Westerterp-Plantenga MS, Lejeune MP, Kovacs EM (2005). Body weight loss and weight maintenance in relation to habitual caffeine intake and green tea supplementation. *Obes Res* **13**, 1195–1204.

Wolfram S, Raederstorff D, Wang Y, Teixeira SR, Elste V, Weber P (2005). TEAVIGO (epigallocatechin gallate) supplementation prevents obesity in rodents by reducing adipose tissue mass. *Ann Nutr Metab* **49**, 54–63.

Yoshioka M, Doucet E, Drapeau V, Dionne I, Tremblay A (2001). Combined effects of red pepper and caffeine consumption on 24 h energy balance in subjects given free access to foods. *Br J Nutr* **85**, 203–211.

Yoshioka M, St-Pierre S, Drapeau V, Dionne I, Doucet E, Suzuki M *et al*. (1999). Effects of red pepper on appetite and energy intake. *Br J Nutr* **82**, 115–123.

Zemel MB (2002). Regulation of adiposity and obesity risk by dietary calcium: mechanisms and implications. *J Am Coll Nutr* **21**, 146S–151S.

Zheng G, Sayama K, Okubo T, Juneja LR, Oguni I (2004). Anti-obesity effects of three major components of green tea, catechins, caffeine and theanine, in mice. *In vivo* **18**, 55–62.