Matcha green tea drinks enhance fat oxidation during brisk walking in females

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Running title: Matcha and exercise-induced fat oxidation

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ABSTRACT
Intake of the catechin epigallocatechin gallate and caffeine has been shown to enhance exercise-induced fat oxidation. Matcha green tea powder contains catechins and caffeine and is consumed as a drink. We examined the effect of Matcha green tea drinks on metabolic, physiological and perceived intensity responses during brisk walking. Thirteen females (age: 27±8 yr, body mass: 65±7 kg, height: 166±6 cm) volunteered. Resting metabolic equivalent (1-MET) was measured using Douglas bags (1-MET: 3.4±0.3 ml·kg⁻¹·min⁻¹). Participants
completed an incremental walking protocol to establish the relationship between walking speed and oxygen uptake and individualize the walking speed at 5- or 6-MET. A randomized cross-over design was used with participants tested between day 9 and 11 of the menstrual cycle (follicular phase). Participants consumed 3 drinks (each drink made with 1 gram of Matcha premium grade, OMGTea Ltd UK) the day before, and 1 drink 2 hours before the 30-min walk at 5- (n=10) or 6-METs (walking speed: 5.8±0.4 km·h⁻¹) with responses measured at 8-10, 18-20 and 28-30 min. Matcha had no effect on physiological and perceived intensity responses. Matcha resulted in lower respiratory exchange ratio (control: 0.84±0.04; Matcha: 0.82±0.04) (P < 0.01) and enhanced fat oxidation during a 30-min brisk walk (control: 0.31±0.10; Matcha: 0.35±0.11 g·min⁻¹) (P < 0.01). Matcha green tea drinking can enhance exercise-induced fat oxidation in females. However, when regular brisk walking with 30-min bouts is being undertaken as part of a weight loss program, the metabolic effects of Matcha should not be overstated.

Key words: catechins; health promotion; treadmill walking

INTRODUCTION

The polyphenol composition of green tea leaves is characterised by the flavonoid catechins i.e. catechin gallate, epicatechin gallate, epigallocatechin gallate, epicatechin epigallate, gallocatechin and gallocatechin gallate (Xu et al., 2004). Due to the processing methods of the leaves, green tea has a high content of catechins compared to oolong and black tea. The green tea components contribute to the antioxidant capacity (Peluso and Serafini, 2017), with epigallocatechin gallate (EGCG) considered the bioactive compound (Khan et al., 2006). The antioxidant capacity of green tea likely contributed to the evidence for health benefits by regular intake of green tea such as a reduced risk for some cancers (Guo et al., 2017) and
cardiovascular and ischemic-related diseases (Pang et al., 2016). Green tea has also been implicated in body-weight management (Janssens et al., 2016) by promoting fat oxidation. EGCG is considered the bioactive compound to promote fat oxidation (Kapoor et al., 2017). Chronic intake of green tea extract enhanced fat oxidation during swimming (Murase et al., 2005) and running in mice (Murase et al., 2006). In addition, EGCG has been shown to reduce body weight in diet-induced obese mice (Lee et al., 2009), indicative of a change in energy balance. In humans, observations on fat oxidation during exercise with short term intake of green tea or EGCG intake were inconsistent. Randell et al (2013) did not observe enhanced fat oxidation during cycling at 50% of maximum power in men after 1 and 7-day intake with no intake on the day of testing. During 2 hr of cycling at 50% of maximum power, green tea extract had no effect on the respiratory exchange ratio (Eichenberger et al., 2010). However, Venables et al (2008) showed enhanced fat oxidation with green tea extract during 30-min cycling exercise at 60% of maximum oxygen uptake in men with the supplement taken on the day before and on the day 1 hr before testing.

In human studies on exercise-induced fat oxidation, the delivery mode of green tea supplementation has been in capsule form. No studies examined the effect of traditional brewed green tea drinks with leaves on fat oxidation during exercise. Matcha green tea powder contains catechins and caffeine and when it is consumed as a drink it ensures an oral intake of all the green tea leaf components. In addition, the process of powdering green tea leaves adds to the potential beneficial effects of Matcha (Fujioka et al., 2016). Therefore, the intake of green tea components by Matcha drinking may be higher than brewed green tea without the leaf consumption and guarantees intake of water-soluble and water-insoluble parts (Xu et al., 2015). In mice fed a high-fat diet, Matcha intake promoted lipid metabolism (Xu et al., 2016). No studies have examined the effect of Matcha drinks on substrate oxidation during exercise in humans.
Regular exercise that is performed to obtain health benefits is recommended to have an exercise intensity between 3 and 6 metabolic equivalents i.e. 3 to 6 times the resting energy expenditure according to physical activity guidelines (Haskell et al., 2007). Walking is a popular physical activity (Paul et al., 2015) and for most people brisk walking meets intensity requirements (Fitzsimons et al., 2005). No studies have examined the effect of a nutritional ergogenic aid on substrate oxidation during brisk walking. Dietary changes and regular exercise may result in a negative energy balance and reduce body weight and body fat. Nutritional ergogenic aids could enhance these effects (Arent et al., 2017). Enhanced fat oxidation during cycling by a decaffeinated green tea extract was associated with a decrease in body fat (Roberts et al., 2015). Fat oxidation during brisk walking with green tea drinking has not been examined. According to Weiss and Anderton (2003), the concentration of EGCG from drinking Matcha green tea is at least 3 times the highest intake of EGCG compared to other green teas.

Therefore, the aim of the present study was to examine the effect of the consumption of Matcha on substrate oxidation, physiological responses and perceived intensity during brisk walking in females.

METHODS

Participants

A randomised, cross-over experimental design was used. Thirteen recreationally active healthy women [age: 27±8 yr, height: 166±6 cm, body mass: 65±7 kg, BMI: 23.5±2.6 kg·m⁻² (range: 19.1-30.2 eleven with 18.9 < BMI < 24.9), means±SD] volunteered and provided written informed consent. All participants were non-smokers. Accepted contraceptive methods were combined pill, diaphragm or intrauterine device. Ethics approval was obtained
Experimental design and preliminary testing

Participants visited the laboratory on three occasions between 9 and 11 o’clock in the morning. During the first visit, height and body mass were measured. Subsequently, participants rested in a chair for 30 minutes with 2 x 10 min expired air collections separated by 5 minutes using the Douglas bag technique to determine the oxygen consumption at rest (i.e. the one metabolic equivalent 1-MET) with the lowest value taken as the 1-MET.

Subsequently, participants completed an incremental-intensity walking test on a treadmill (HP Cosmos Pulsar Bodycare Products UK) with 5 x 8-min stages. Starting speed was 2 km∙h⁻¹ with a stage increment of 1 km∙h⁻¹ until speed reached 6 km∙h⁻¹. During each stage, expired air was collected in the last 3 minutes. The incremental-intensity walking test was performed to determine the linear relationship between walking speed and oxygen consumption expressed as the metabolic equivalent. For each individual, the linear relationship between walking speed and metabolic equivalent (r² = 0.9353±0.0383) was used to calculate the walking speed at 5- or 6-METs (i.e. moderate intensity exercise). For visits two and three with either having Matcha or no supplement, participants were tested in the follicular phase of the menstrual cycle (i.e. 9-11 days following start of the menstruation).

Hormonal levels were not measured and determination of the follicular phase was based on verbal information provided by the participants. In preparation for all testing sessions, participants abstained from strenuous and unaccustomed exercise for 48 hours, no alcohol for 24 hours before testing and no caffeine-containing products on the day of testing.
For the Matcha condition, participants consumed 3 x 1 gram of Matcha powder (Matcha premium grade OMG Tea Ltd, UK) mixed with water at meal times on the day before testing. On the day of testing, participants consumed 1 gram of Matcha with water two hours before arrival and arrived following an overnight fast. The supplementation strategy was based on Venables et al (2008). One gram of Matcha premium grade contains 143 mg total catechins and 30 mg caffeine (composition data from OMG Tea Ltd, UK). Before visit two, participants recorded their dietary intake for 48 hours and were instructed to match the same dietary intake 48 hours before arrival for visit three. The intake before visit three was recorded on a new food diary. Carbohydrate fat and protein intake and total energy intake (kJ) were quantified with Nutritics (Nutritics LTD Dublin Ireland).

Participants walked on a treadmill at a speed to elicit 5- or 6-MET for 30 minutes with expired air collected from 8 to 10, 18 to 20 and 28 to 30 minutes with recording of heart rate (Polar Vantage NV Polar Electro Oy Kempele Finland) and rating of perceived exertion (Borg 6 to 20 scale) (Borg, 1982). Expired air was analyzed with a three-point calibrated gas analyser (Servomex Series 1400 gas analyser Servomex Crowborough United Kingdom) and volume measured (Harvard Apparatus Ltd. Edenbridge United Kingdom). Gas volumes were corrected to standard temperature and pressure and dry gas conditions (STPD) and calculated using Haldane transformation with consideration of inspired fractions of oxygen and carbon dioxide at the time of expired air collection. Rates of whole body fat and carbohydrate oxidation were calculated with equations 1 and 2 from Jeukendrup and Wallis (2005) and the assumption of negligible protein oxidation:

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Fat\ oxidation\ (g \cdot min^{-1}) = 1.695 \times \dot{V}O_2 - 1.701 \times \dot{V}CO_2
\]  \hspace{1cm} (1)

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Carbohydrate\ oxidation\ (g \cdot min^{-1}) = 4.210 \times \dot{V}CO_2 - 2.962 \times \dot{V}O_2
\]  \hspace{1cm} (2)
Statistical analysis

Analyses were completed using Graphpad Prism version 5.00 for Window (GraphPad Software San Diego California USA). A power analysis indicated that a sample size of 13 was required to allow a detection of a 15% increase in fat oxidation from a baseline value of fat oxidation of 0.25 g·min⁻¹ (Dasilva et al., 2011) with a SD of 0.07 for both groups with a high statistical power (1−β = 0.80; 0.05 = α level). A two-way ANOVA was used to analyse oxygen consumption, carbon dioxide production and substrate oxidation for time effects with post-hoc paired samples t-tests. Means were calculated for all parameter values collected from 8 to 10, 18 to 20 and 28 to 30 minutes during the 30-min treadmill walk. Data normality was assessed with D'Agostino-Pearson normality tests. Paired samples t-tests were conducted to compare parameter values between control and Matcha conditions. Statistical significance was accepted at P < 0.05.

RESULTS

The 1-MET was 3.4±0.3 mL·kg⁻¹·min⁻¹, means±SD range: 2.9–3.8 mL·kg⁻¹·min⁻¹). There were no differences in absolute values of daily dietary intake parameters (i.e. carbohydrate, control: 182±71 g, Matcha: 157±49 g; fat, control: 67±23 g, Matcha: 72±22 g, protein, control: 70±43 g, Matcha: 75±35 g; total energy intake, control: 6697±2302 kJ, Matcha: 6604±1796 kJ). Participants were low caffeine consumers (control: 48±57 mg, Matcha: 40±45 mg).

Matcha vs control

Physiological responses and rating of perceived exertion
Participants walked in the control and Matcha condition at an individualized walking speed for 30 minutes with an exercise intensity of 5- or 6-MET. Ten participants walked at 5-MET (walking speed: 5.7±0.4 km·h⁻¹) to avoid those participants willing to jog at the treadmill speed of 6-MET. For the three participants walking at 6-MET, the walking speed was 6.0±0.5 km·h⁻¹. Oxygen uptake (control: 18.1±2.8; Matcha: 18.1±2.8 mL·kg⁻¹·min⁻¹), minute ventilation (control: 25.9±3.3; Matcha: 25.2±3.3 L·min⁻¹) and heart rate (control: 119±18; Matcha: 120±17 beats·min⁻¹) were not different. Figure 1 shows the oxygen (A) and carbon dioxide (B) values over time with no time effects. Rating of perceived exertion during walking at an intensity of 5- or 6-MET was not different compared to the Matcha condition (control: 11±1; Matcha: 12±1).

Respiratory exchange ratio and substrate oxidation

Figure 2 shows substrate oxidation as a function of time during the 30-min walk. Time effects for carbohydrate oxidation showed a trend to be lower at 28-30 min compared to 8-10 min in the placebo condition (P = 0.07) and lower in the Matcha condition (P = 0.01) (Figure 2A). In the placebo condition, there was a trend for fat oxidation at 28-30 min to be higher than fat oxidation at 8-10 min (P = 0.06) (Figure 2B). Fat oxidation at 28-30 min was higher than fat oxidation at 18-20 min (P = 0.04) (Figure 2B). In the Matcha condition, fat oxidation at 28-30 min was higher than fat oxidation at 8-10 min (P = 0.04) and 18-20 min (P < 0.01) (Figure 2B). The respiratory exchange ratio was 0.02 units lower in the Matcha condition (Figure 3) indicating a larger contribution of fat as an energy source. In the Matcha condition, carbohydrate oxidation was lower (control: 0.69±0.18; Matcha: 0.56±0.20 g·min⁻¹) (Figure 4) and fat oxidation was higher (control: 0.31±0.10; Matcha: 0.35±0.11 g·min⁻¹) (Figure 5) over the full 30-min of the walk. The individual observations on carbohydrate (Figure 4) (i.e. 11 participants lower values) and fat oxidation (Figure 5) (i.e. 10 participants higher values)
seem to indicate that the absolute changes in substrate oxidation were not related to the values observed in the control condition.

4. Discussion

With Matcha green tea drinking, polyphenol and caffeine intake occurs by whole consumption of the powdered green tea leaves. Previous studies on the effects of green tea on exercise responses used capsulated intake of green tea extract or EGCG (Dean et al., 2009; Eichenberger et al., 2009; Martin et al., 2014; Venables et al., 2008) or enriched canned drinks with green tea catechins and caffeine (Hodgson et al., 2013; Randell et al., 2013). We are not aware of studies on the effects on exercise responses by traditionally brewed green tea drinking by which the intake of catechines and caffeine is not by the consumption of green tea leaves. Females in our study consumed 4 normal cups of Matcha green tea in 24 hours. We observed an enhanced fat oxidation with Matcha green tea drinking during 30 min of brisk walking in females. Our observation is similar to that in a study by Venables et al (2008) with effects of green tea extract in enhancing fat oxidation during 30-min cycling at 60% VO$_{2\text{max}}$ in males. In the study by Venables et al (2008) participants were dosed 2 times the day before and 1 h before testing with a green tea extract that contained in total 890 mg of polyphenols and 408 mg EGCG but was without caffeine. EGCG intake in the present study with 4 cups of Matcha green tea over a 24 hour period amounted to a total intake of 292 mg EGCG and 120 mg caffeine. The final intake in the present study provided 73 mg of EGCG and 30 mg of caffeine whereas the dose in Venables et al (2008) was 86% higher i.e. 136 mg EGCG but no caffeine. It is possible that the components of Matcha provide a synergistic effect on exercise-induced fat oxidation. A comparison with other studies on the effects of the intake of green tea extract or EGCG is problematic due to differences in dosing strategy i.e. amounts, intake duration, intake composition, training status of participants and fed or fasted
status testing. For example, Martin et al. (2014) did not observe an effect of green tea during exercise but as participants were provided with a standardized breakfast 90 min before the exercise test, this may have affected the observed substrate oxidation during the exercise. Eichenberger et al. (2009) examined green tea extract effects during 2 hr cycling in endurance trained men cycling > 6 hours per week, and the absence of a green tea effect could be due to training status of the subjects. However, observations of enhanced fat oxidation in the present study seem to indicate that it is possible to have intake by a cup of Matcha of essential catechins e.g. EGCG and caffeine in amounts that cannot be achieved with a cup of traditional brewed green tea. The caffeine intake in our study was very small: a total of 120 mg over 24 hours. An acute intake of 6 mg/kg of body mass of caffeine reduced the respiratory exchange ratio during exercise (Cruz et al., 2015). In the present study, the intake of 30 mg of caffeine by Matcha intake on the day of testing was less than 0.5 mg/kg of body mass, an amount for which there is no evidence for affecting exercise-induced fat oxidation.

EGCG is considered the bioactive compound in green tea acting by inhibition of catechol-O-methyltransferase (i.e. COMT). In general, inhibition of COMT would reduce the breakdown of catecholamines and promote an internal cellular environment for enhanced fat oxidation. However, a study by Hodgson et al. (2013) observed that 8-day intake of green tea extract did not enhance adrenergic stimulation during exercise. Therefore, due to the absence of differences in the adrenergic system with intake of green tea extract in-vivo, the inhibition of COMT may not cause the observed enhanced fat oxidation. Alternatively, EGCG has been linked with activation of the transient receptor potential vanilloid type 1 (i.e. TRPV1) and eNOS activation (Guo et al., 2015). In addition, TRPV1 is also linked with eNOS activation and NO production (Yu et al., 2017). Activation of eNOS would result in increased production of nitric oxide and enhanced blood flow with improved delivery of free fatty acids. Interestingly, activation of TRPV1 was associated with enhanced fat oxidation in male
mice by capsiate supplementation (Haramizu et al., 2006), possible by contributing to functional sympatholysis during exercise (Ives et al., 2017). However, differences in both the metabolite profiles in bioavailability of plasma catechins in animal studies and the amount of catechins used to examine effects in endothelial cells (Guo et al., 2015), warrants caution to generalize from these findings to observations with green tea extracts or powder in humans.

The exercise modality in the present study was walking in females with an intensity known to provide health benefits. In addition, females were tested during the follicular phase but evidence on hormonal effects on fat oxidation in females during exercise is inconsistent (Kaneley et al., 1992; Vaiksaar et al., 2011; Wenz et al., 1997). In addition, we had no objective measurement of the follicular phase by hormonal observations and cannot exclude that the variation in individual responses may be due to intra-individual differences in hormonal levels. We also did not control the physical activity status of the participants.

Future studies may want to examine the effects of Matcha green tea drinking for longer duration and combined with an exercise intervention in normal weight overweight and obese individuals. In addition, future studies on nutritional interventions that enhance fat oxidation during exercise should address the causality of high responders. It is of interest also to explore in future studies whether enhanced fat oxidation by Matcha green tea would affect insulin sensitivity. A study by Robinson et al (2015) observed that maximal fat oxidation during exercise was associated with insulin sensitivity.

In summary, Matcha green tea drinking, just 4 cups in 24 hours enhanced fat oxidation during brisk walking in healthy females. The composition of Matcha green tea leaves is sufficient for habitual Matcha drinking to provide beneficial metabolic responses during brisk walking. However, when regular moderate intensity exercise is undertaken as part of a weight loss program, the effects of Matcha should not be overstated.
Acknowledgments: Matcha premium green tea powder was provided by OMGTea Ltd (United Kingdom).

REFERENCES


**Figure legends**

**Figure 1.** Oxygen consumption (A) and carbon dioxide production (B) at different time points during the 30-min treadmill walk individualized walking speeds at 5-MET (10 participants) or 6-MET (3 participants). Data are presented as mean ± SEM.

**Figure 2.** Carbohydrate oxidation (A) and fat oxidation (B) in the control and Matcha condition at different time points during a 30-min treadmill walk at individualized walking speeds at 5-MET (10 participants) or 6-MET (3 participants). A, * different between time points 28-30 and 8-10 min in the Matcha condition. B, $ different between time points 28-30 min and 8-10 in the control condition and * different between 28-30 min and 8-10 min and 18-20 min in the Matcha condition.

**Figure 3.** Respiratory exchange ratio (RER) in the control and Matcha condition during a 30-min treadmill walk at individualized walking speeds at 5-MET (10 participants) or 6-MET (3 participants). Data are presented as mean ± SD. *, difference between conditions.

**Figure 4.** Carbohydrate oxidation in the control and Matcha condition during a 30-min treadmill walk at individualized walking speeds at 5-MET (10 participants) or 6-MET (3 participants). Columns represent mean values. * different between conditions.
Figure 5. Fat oxidation in the control and Matcha condition during a 30-min treadmill walk at individualized walking speeds at 5-MET (10 participants) or 6-MET (3 participants). Columns represent mean values. * different between conditions.