

## Acute ingestion of catechin-rich green tea improves postprandial glucose status and increases serum thioredoxin concentrations in postmenopausal women

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### Abstract

Elevated postprandial hyperglycaemia and oxidative stress increase the risks of type 2 diabetes and CVD. Green tea catechin possesses antidiabetic properties and antioxidant capacity. In the present study, we examined the acute and continuous effects of ingestion of catechin-rich green tea on postprandial hyperglycaemia and oxidative stress in healthy postmenopausal women. Participants were randomly assigned into the placebo (P, *n* 11) or green tea (GT, *n* 11) group. The GT group consumed a catechin-rich green tea (catechins 615 mg/350 ml) beverage per d for 4 weeks. The P group consumed a placebo (catechins 92 mg/350 ml) beverage per d for 4 weeks. At baseline and after 4 weeks, participants of each group consumed their designated beverages with breakfast and consumed lunch 3 h after breakfast. Venous blood samples were collected in the fasted state (0 h) and at 2, 4 and 6 h after breakfast. Postprandial glucose concentrations were 3% lower in the GT group than in the P group (three-factor ANOVA, group × time interaction, *P* < 0.05). Serum concentrations of the derivatives of reactive oxygen metabolites increased after meals (*P* < 0.05), but no effect of catechin-rich green tea intake was observed. Conversely, serum postprandial thioredoxin concentrations were 5% higher in the GT group than in the P group (three-factor ANOVA, group × time interaction, *P* < 0.05). These findings indicate that an acute ingestion of catechin-rich green tea has beneficial effects on postprandial glucose and redox homeostasis in postmenopausal women.

**Key words:** Catechins: Postprandial glucose: Postprandial oxidative stress: Postprandial antioxidant capacity

Green tea polyphenols, catechins in particular, have beneficial effects that prevent the development of diabetes and CVD. In fact, some epidemiological studies have reported that regular green tea consumption is associated with reduced CVD-related mortality<sup>(1–3)</sup>. Although a previous meta-analysis has found that regular consumption of green tea was associated with a significant reduction in fasting glucose<sup>(4)</sup> and insulin concentrations<sup>(5)</sup>, limited evidence is available regarding the effect of green tea on postprandial hyperglycaemia, which is an independent risk factor for CVD and type 2 diabetes mellitus<sup>(6,7)</sup>. Therefore, it is necessary to examine the effects of green tea consumption on postprandial hyperglycaemia.

Human studies have demonstrated that postprandial hyperglycaemia and hyperlipidaemia are positively correlated with oxidant production<sup>(8–10)</sup>. Moreover, a previous study<sup>(11)</sup>

has shown that intake of a high-lipid diet resulted in greater postprandial oxidative stress than intake of a carbohydrate diet with the same energy content. Elevated postprandial oxidative stress plays a role in the development of atherosclerosis through endothelial dysfunction<sup>(12,13)</sup>. Thus, antioxidant supplementation may be key in preventing postprandial oxidative stress.

Many investigations have highlighted the effects of antioxidant supplementation, including green tea, on postprandial oxidative stress<sup>(13–15)</sup>. The antioxidant effects of green tea are partly attributed to catechins. Some studies<sup>(16–18)</sup> have reported that green tea polyphenols, specifically catechins, ameliorate the elevation of fasting oxidative stress. However, as there is no conclusive evidence for the effect of green tea on postprandial oxidative stress, further research is needed

**Abbreviations:** d-ROM, derivatives of reactive oxygen metabolites; GT, green tea; P, placebo; TRX, thioredoxin.

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to determine the role of catechins on postprandial oxidative stress in postmenopausal women who are likely to show increased oxidative stress<sup>(19)</sup>. To date, no information is available regarding the effect of green tea on postprandial oxidative stress in postmenopausal women. From the viewpoint of ageing and menopause, interventions that modulate oxidative stress are needed to examine this age group. The purpose of the present study was to examine the acute and continuous effects of the ingestion of catechin-rich green tea on postprandial glucose and oxidative stress in postmenopausal women. We hypothesised that consumption of catechin-rich green tea would ameliorate postprandial hyperglycaemia and oxidative stress.

## Methods

### Participants

The present study was conducted according to the guidelines laid out in the Declaration of Helsinki, and all procedures involving human participants were approved by the Research Ethics Committees of the Tokyo Gakugei University (trial no. 2012-57). Written informed consent was obtained from all participants. The study participants consisted of Japanese postmenopausal women aged 62–73 years ( $n$  22) who were randomly recruited from the general population of local communities. All the included participants had no difficulties with mobility or performing their daily activities. Participants with a history of CVD, those who used glucose-lowering medication in the previous 3 months, smokers, or women <60 years of age were excluded from the study. For the baseline evaluation, we administered a simple lifestyle-related questionnaire (physical activity, medication, sleep, alcohol intake and smoking). Participants were asked to maintain their daily diet and lifestyle during the study period.

### Anthropometry

Body mass was measured to the nearest 0.1 kg using a digital scale (InnerScan 50; Tanita Corporation). Height was measured to the nearest 0.1 cm using a wall-mounted stadiometer (YS-OA; As One Corporation). BMI was calculated as weight (kg) divided by the square of height (m<sup>2</sup>). Waist circumference was measured to the nearest 0.1 cm at the level of the umbilicus using a flexible plastic tape. Arterial blood pressure was measured from the right arm of the participants while in a seated position by using a standard mercury sphygmomanometer (605P; Yagami Company Limited). Participants were seated in a chair for 5 min before blood pressure was measured. At least two measurements were taken at each time point, and the mean of these values was recorded.

### Green tea and placebo beverage content

Green tea and placebo beverages used in the present study were provided by Kao Corporation. A brewed green tea with a natural flavour was used as the placebo beverage,

and catechin-rich green tea was prepared with a green tea extract and hot water. The green tea beverage contained 615 mg/350 ml of total catechins (33.2 mg catechin, 22.9 mg catechin gallate, 135.2 mg gallic catechin, 108.4 mg gallic catechin gallate, 34.8 mg epicatechin, 39.9 mg epicatechin gallate, 114.5 mg epigallocatechin and 125.9 mg epigallocatechin gallate) and 77.0 mg caffeine. The placebo beverage contained 92 mg/350 ml of total catechins (5.7 mg catechin, 3.0 mg catechin gallate, 28.6 mg gallic catechin, 18.0 mg gallic catechin gallate, 4.1 mg epicatechin, 3.8 mg epicatechin gallate, 14.2 mg epigallocatechin and 15.0 mg epigallocatechin gallate) and 85.2 mg caffeine. The green tea and placebo beverages were adjusted to contain the same levels of vitamin C and other minor polyphenols.

### Experimental protocol

A double-blind, placebo-controlled, parallel design was used. The twenty-two postmenopausal women were randomly divided into the following two groups: placebo (P,  $n$  11) group and green tea (GT,  $n$  11) group. The physical characteristics of both groups are presented in Table 1. The participants of the GT and P groups were instructed to consume one bottle of their respective beverage per d with their breakfast for 4 weeks. Previous studies<sup>(20,21)</sup> have reported that a 4-week period of green tea ingestion can reduce fasting circulating concentrations of oxidative stress markers such as oxidised LDL and malondialdehyde.

At baseline and after 4 weeks, all participants visited the laboratory at 08.30 hours after a 10 h overnight fast (no intake of food or drink, except water). After a 15 min rest, a fasting venous blood sample was collected by venepuncture while participants were in a seated position. Participants then consumed a standardised breakfast. A clock was started when the participants began eating, and they were required to rest (e.g. read or write) in the laboratory for 6 h after the initiation of the breakfast meal. A second test meal (identical to the first meal) was consumed 3 h after the initiation of the first meal. Further venous blood samples were collected at 2, 4 and 6 h after the initiation of the breakfast meal.

### Test meals

The test meals consisted of white bread, sliced cheese, yogurt, ham, mayonnaise, lettuce, tomato, butter and peanut butter. The meal was prescribed according to body mass; hence, we provided 1.00 g fat, 1.24 g carbohydrates, 0.52 g protein and 67 kJ energy per kg body mass. Energy derived was 56.3% from fat, 30.8% from carbohydrate and 12.9% from protein. Previous studies<sup>(22,23)</sup> have reported that this macronutrient composition increased postprandial oxidative stress. All participants were asked to consume the test meal within 20 min. The time taken to consume the first postprandial test meal (i.e. at baseline) was recorded and replicated in the subsequent postprandial test meal (i.e. at week 4). None of the participants reported nausea or any gastrointestinal discomfort during or after each meal. Participants consumed either a placebo (catechins 92 mg/350 ml) or green tea (catechins

**Table 1.** Changes in physical characteristics, green tea and coffee consumption at baseline and after 4 weeks (Mean values with their standard errors)

	Placebo group (n 11)		Green tea group (n 11)		P (time)	P (interaction)
	Mean	SE	Mean	SE		
Age (years)						
Baseline	66.5	0.6	66.6	1.2	–	–
4 weeks						
Height (m)						
Baseline	1.53	0.01	1.53	0.02	–	–
4 weeks						
Body mass (kg)						
Baseline	55.4	2.4	55.7	2.0	0.001	0.843
4 weeks	53.9	2.3	54.1	1.8		
BMI (kg/m <sup>2</sup> )						
Baseline	23.4	0.8	23.8	0.8	0.001	0.811
4 weeks	22.7	0.8	23.1	0.8		
Waist circumference (cm)						
Baseline	84.7	2.3	83.7	2.4	0.610	0.581
4 weeks	84.8	2.4	82.3	2.2		
Systolic blood pressure (mmHg)						
Baseline	128	4	125	5	0.665	0.899
4 weeks	126	3	124	3		
Diastolic blood pressure (mmHg)						
Baseline	81	2	77	2	0.001	0.652
4 weeks	76	1	71	2		
Green tea consumption (cups/d)						
Baseline	4.4	0.4	3.3	0.3	0.679	0.492
4 weeks	3.9	0.4	3.7	0.3		
Coffee consumption (cups/d)						
Baseline	1.6	0.2	2.0	0.3	0.100	0.566
4 weeks	2.0	0.2	1.3	0.1		

615 mg/350 ml) beverage with breakfast. Participants consumed water *ad libitum* during the first postprandial testing, and the pattern and volume ingested was replicated in the subsequent postprandial testing.

#### Standardisation of the diet and physical activity

All participants weighed and recorded all food and drink consumed during the day before each postprandial testing period (i.e. at baseline and at week 4). All participants refrained from drinking alcohol 2 d before each postprandial testing, and they replicated their dietary intake during the day before each postprandial testing. Additionally, all participants were asked to remain inactive on the day before each postprandial testing and throughout each postprandial testing. For the determination of physical activity levels, all participants were instructed to wear a uniaxial accelerometer (Lifecorder EX; Suzuken Company Limited) during the experimental period (i.e. at 4 weeks). A number of studies<sup>(24,25)</sup> have reported that the Lifecorder EX is considered as a validated accelerometer for evaluating and monitoring physical activity levels.

#### Blood collection and analysis

For measuring serum blood markers, samples were allowed to clot for 30 min at room temperature and then centrifuged at

1300 g for 10 min at 4°C. The serum sample was dispensed into plain microtubes and stored at –80°C until the assay. For measuring plasma blood markers, blood samples collected into tubes containing EDTA were immediately centrifuged and stored at –80°C until the assay. Plasma concentrations of total catechins, gallic catechin, gallic catechin gallate, epigallocatechin gallate, epigallocatechin and epigallocatechin gallate were measured by using HPLC with solid-phase extraction as described previously<sup>(26,27)</sup>. Enzymatic colorimetric assays were used to measure the plasma concentrations of glucose (GLU-HK (M); Shino-Test Corporation), serum TAG (Pure-Auto S TG-N; Sekisui Medical Company Limited) and NEFA (NEFA-HR; Wako Pure Chemical Industries Limited). Plasma concentrations of insulin were measured by ELISA. Serum concentrations of the derivatives of reactive oxygen metabolites (d-ROM) and biological antioxidant potential were measured using assay kits (Diacron). The d-ROM test measures the oxidative stress of blood samples by evaluating the level of reactive oxygen metabolites. This assay is based on the capability of *N,N*-diethyl-*p*-phenylenediamine to give a stable, coloured solution when it is transformed into its radical cation (*N,N*-diethyl-*p*-phenylenediamine<sup>+</sup>)<sup>(28)</sup>. The biological antioxidant potential assay is a photometric test that determines the serum concentration of antioxidants capable of reducing Fe from the ferric to the ferrous form<sup>(29)</sup>. Plasma H<sub>2</sub>O<sub>2</sub> concentration was measured using the Amplex Red reagent method (Molecular Probes, Invitrogen

Detection Technologies). Concentrations of plasma thioredoxin (TRX; Immuno-Biological Laboratories Company Limited) were measured by ELISA.

**Statistical analysis**

Data were analysed using Predictive Analytics Software, version 18.0 for Windows (SPSS, Inc., IBM). Based on previous studies<sup>(30,31)</sup>, we estimated the sample size of the present study (control (*n* 11) and exercise (*n* 11) groups). In fact, power analysis indicated that eleven subjects per group were required to obtain a large effect size (multi-way ANOVA, >0.80) with an 80% power at  $\alpha = 0.05$ . The effect size was calculated using the method outlined by Cohen<sup>(32)</sup>. The Kolmogorov–Smirnov test was used to check for the normality of the distribution of all the blood parameters. The distribution of these parameters did not differ significantly from normal values. Unpaired Student's *t* tests were used to assess group differences between data acquired at baseline and at 4 weeks. Three-factor ANOVA with repeated measures was used to determine the effect of group (P or GT), intervention (baseline or week 4), and postprandial interval (0–6 h) on the concentrations of blood markers. When significant interaction effects were detected, simple main-effects tests

were employed. Statistical significance was accepted at the 5% level ( $P < 0.05$ ). Results are presented as means with their standard errors.

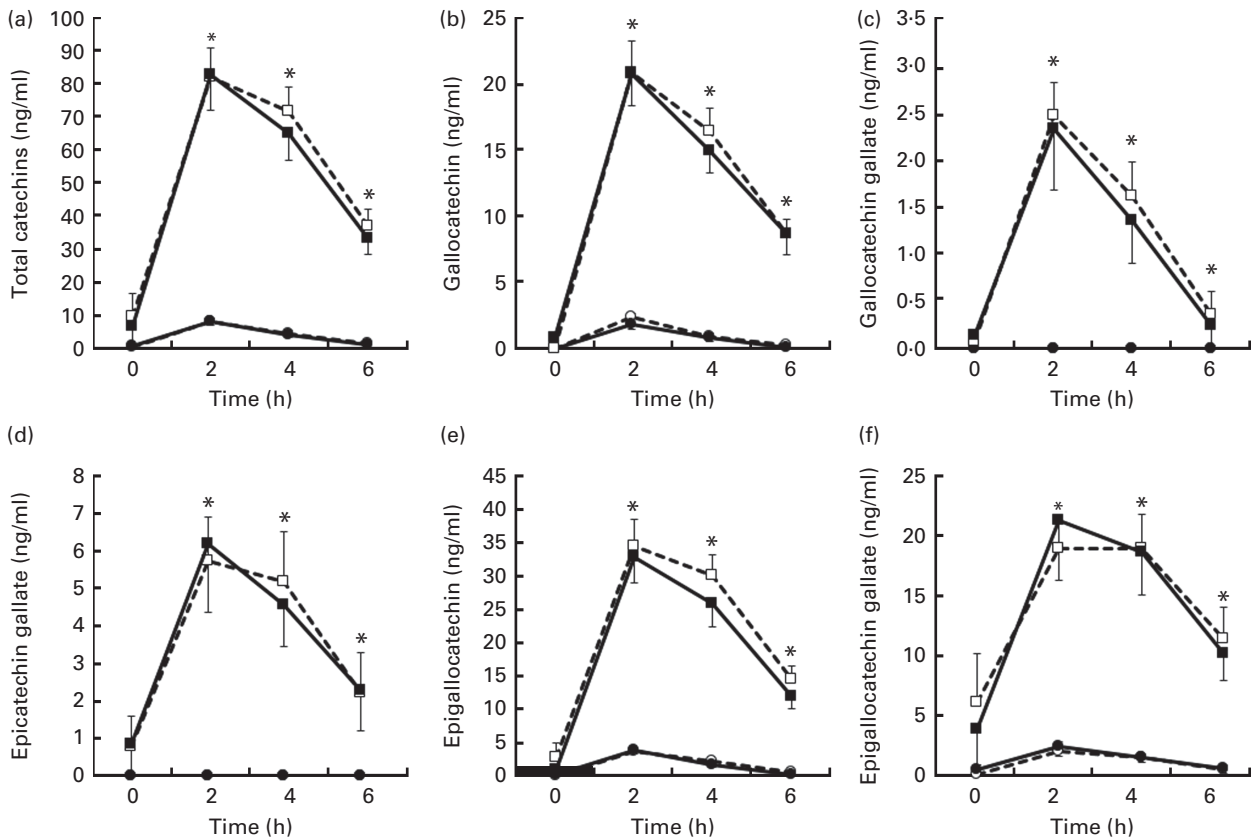
**Results**

*Physiological characteristics and physical activity*

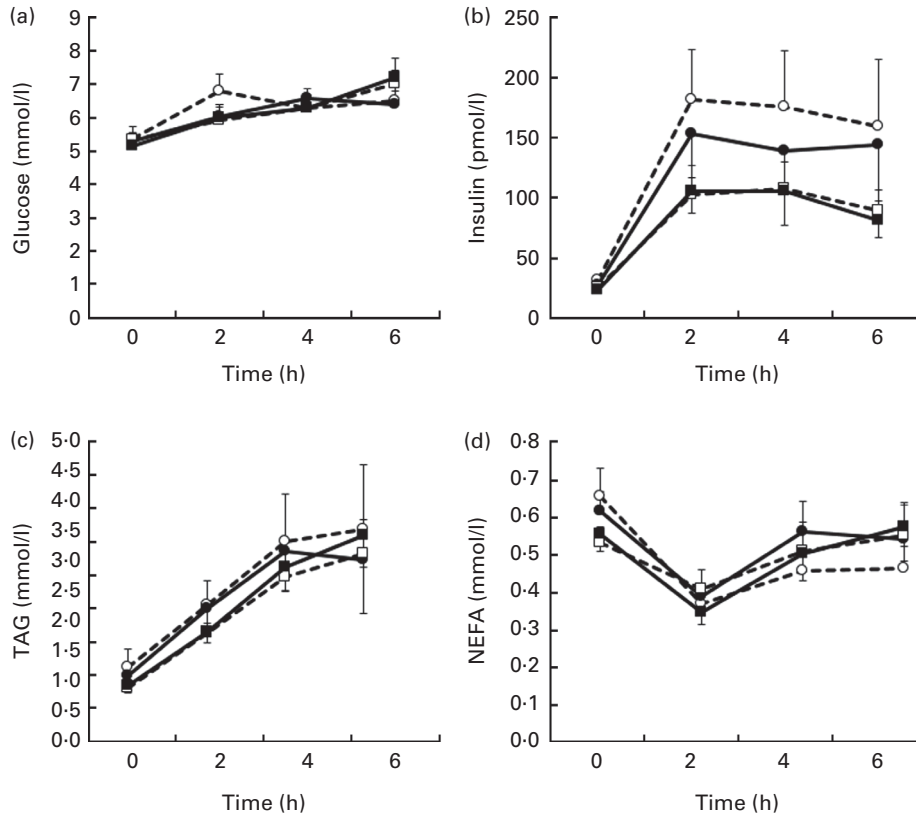
There were no differences in the physical and physiological characteristics between the groups at baseline and at week 4 (Table 1). No change was observed in the daily activity habits of the participants during the monitoring period (i.e. moderate to vigorous physical activity and step counts). During the study period, the moderate-to-vigorous physical activity level was 172.4 (SE 13.1) and 203.6 (SE 21.7) min/week for the P and GT groups, respectively. The step counts were 7627 (SE 594) and 8226 (SE 633) steps/d for the P and GT groups, respectively. Both moderate-to-physical activity level and step counts did not differ between the groups.

*Fasting and postprandial catechins*

Plasma concentrations of total catechins, gallo catechin, gallo catechin gallate, epicatechin gallate, epigallocatechin



**Fig. 1.** Fasting and postprandial plasma concentrations of total catechins (a), gallo catechin (b), gallo catechin gallate (c), epicatechin gallate (d), epigallocatechin (e) and epigallocatechin gallate (f) in the placebo (P) and green tea (GT) groups at baseline and week 4. —○—, P baseline; —●—, P week 4; —□—, GT baseline; —■—, GT week 4. Values are means, with their standard errors represented by vertical bars. \* Mean value was significantly different from that of the P group ( $P = 0.001$ ; three-factor ANOVA). There were significant time ( $P = 0.001$ ), group ( $P = 0.001$ ) and time  $\times$  group interaction effects ( $P = 0.001$ ) observed for all the individual catechins (three-factor ANOVA).



**Fig. 2.** Fasting and postprandial concentrations of plasma glucose (a), plasma insulin (b), serum TAG (c) and serum NEFA (d) in the placebo (P) and green tea (GT) groups at baseline and week 4. —○—, P baseline; —●—, P week 4; —□—, GT baseline; —■—, GT week 4. Values are means, with their standard errors represented by vertical bars. There was a significant time effect ( $P=0.001$ ) observed for glucose, insulin, TAG and NEFA concentrations, and the time  $\times$  group interaction was significant ( $P=0.013$ ) for glucose concentrations (three-factor ANOVA).

and epigallocatechin gallate are shown in Fig. 1(a)–(f). At the baseline level, there was no difference in all the individual catechins in the plasma between the groups. For all the individual catechins in the plasma, three-factor ANOVA with repeated measures showed a significant main effect of time ( $P=0.001$ ) and group ( $P=0.001$ ). Also, a significant time  $\times$  group interaction ( $P=0.001$ ) was found for all the individual catechins in the plasma. The simple main-effects tests revealed that the concentrations of total catechins, gallic acid, epigallocatechin gallate, epigallocatechin and epigallocatechin gallate were higher in the GT group than in the P group at 2, 4 and 6 h after consumption of the meals (all time points;  $P=0.001$ ). The simple main-effects tests also revealed that gallic acid concentrations were higher in the GT group than in the P group at 2 and 4 h after consumption of the meals ( $P=0.001$ ). The peak concentrations of all the individual catechins were observed at 2 h after consumption of the green tea beverage in the GT group. In addition, all the individual catechin concentrations did not significantly change in either group after 4 weeks compared with the baseline.

#### Fasting and postprandial metabolites/hormones

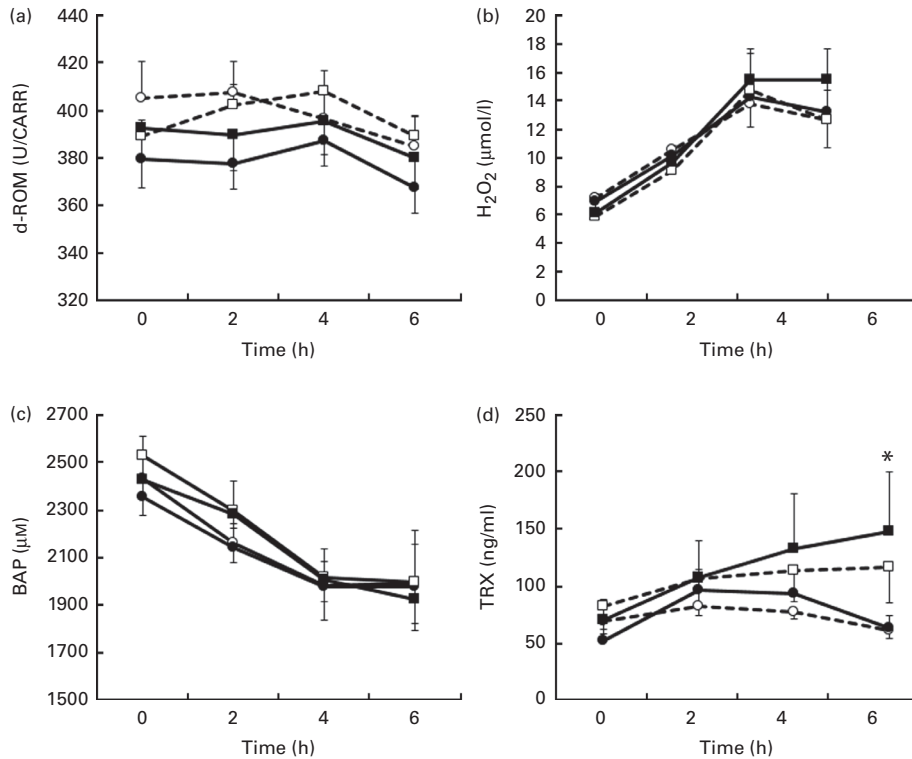
There were no differences in the values of all the blood parameters between the groups at baseline. Plasma glucose concentrations are shown in Fig. 2(a). Three-factor ANOVA

with repeated measures showed a significant main effect of time ( $P=0.001$ ) for plasma glucose concentrations. Significant interactions between time and group ( $P=0.013$ ) were also found. Conversely, plasma glucose concentrations did not significantly change in either group after 4 weeks compared with baseline. Plasma insulin concentrations are shown in Fig. 2(b). For plasma insulin concentrations, three-factor ANOVA with repeated measures showed a significant main effect of time ( $P=0.001$ ) and a tendency of a main effect of group ( $P=0.055$ ). For serum TAG and NEFA concentrations, three-factor ANOVA with repeated measures showed a significant main effect of time ( $P=0.001$ ) (Fig. 2(c) and (d)).

#### Fasting and postprandial oxidative stress markers

Serum d-ROM concentrations are shown in Fig. 3(a). Three-factor ANOVA with repeated measures showed a significant main effect of time ( $P=0.001$ ) for serum d-ROM concentrations. For plasma  $H_2O_2$  and serum biological antioxidant potential concentrations, three-factor ANOVA with repeated measures showed a significant main effect of time ( $P=0.001$ ; Fig. 3(b) and (c)). Serum TRX concentrations are shown in Fig. 3(d). For serum TRX concentrations, three-factor ANOVA with repeated measures showed a significant main effect of time ( $P=0.001$ ) and a time  $\times$  group interaction ( $P=0.012$ ). The simple main-effects tests revealed that serum





**Fig. 3.** Fasting and postprandial concentrations of serum derivatives of reactive oxygen metabolites (d-ROM; Carratelli units (U/CARR)) (a), plasma hydrogen peroxide (b), serum biological antioxidant potential (BAP) (c) and plasma thioredoxin (TRX) (d) in the placebo (P) and green tea (GT) groups at baseline and week 4. -○-, P baseline; -●-, P week 4; -□-, GT baseline; -■-, GT week 4. Values are means, with their standard errors represented by vertical bars. \* Mean value was significantly different from that of the P group ( $P=0.030$ ; three-factor ANOVA). There was a significant time effect ( $P=0.001$ ) observed for d-ROM, hydrogen peroxide, BAP and TRX concentrations, and the time  $\times$  group interaction was significant ( $P=0.012$ ) for TRX concentrations (three-factor ANOVA).

TRX concentrations were higher in the GT group than in the P group at 6 h after consumption of the meals ( $P=0.030$ ). However, serum TRX concentrations did not significantly change in either group after 4 weeks compared with baseline.

## Discussion

To the best of our knowledge, the present study is the first to examine the effects of green tea intake on postprandial glucose and oxidative stress in postmenopausal women. The main finding of the present study is that an acute ingestion of catechin-rich green tea reduced postprandial plasma glucose concentrations in postmenopausal women. We also observed that an acute ingestion of catechin-rich green tea increased postprandial TRX concentrations. The present findings indicate that the intake of catechin-rich green tea improves postprandial hyperglycaemia and redox homeostasis in postmenopausal women. Conversely, there were no continuous effects associated with the ingestion of catechin-rich green tea on postprandial hyperglycaemia and oxidative stress.

Green tea is a rich source of polyphenol antioxidants called catechins, mainly epigallocatechin gallate, epigallocatechin, epicatechin gallate and epicatechin. Several studies<sup>(4,5,16,33)</sup> have reported that green tea catechins possess antidiabetic properties and antioxidant capacity. However, few studies have measured blood catechin concentrations after drinking

green tea for the evaluation of its antidiabetic and antioxidant effects. Thus, a major strength of the present study is that we directly measured blood catechin concentrations in response to an acute ingestion of catechin-rich green tea, and found a significant elevation in the plasma concentrations of total catechins, gallic acid, gallic acid gallate, epigallocatechin gallate, epigallocatechin and epigallocatechin gallate. Moreover, the peak concentration of all the individual catechins was observed at 2 h after ingestion of the green tea beverage in the GT group. These findings support a previous investigation where plasma catechin concentrations were elevated within 1–2 h after intake of green and black tea<sup>(34)</sup>.

In the present study, catechin-rich green tea attenuated postprandial plasma glucose concentrations in postmenopausal women. Moreover, postprandial insulin concentrations had a tendency to be lower in the GT group than in the P group. A previous investigation showed that the acute ingestion of green tea extract improved insulin sensitivity and glucose tolerance in young men<sup>(31)</sup>. Our data in postmenopausal women are consistent with that study. Green tea catechins have been reported to increase adipocyte insulin receptor binding and membrane GLUT4 protein content in rats<sup>(35,36)</sup>. Another study has shown that epigallocatechin gallate enhances the expression of genes associated with insulin sensitivity<sup>(37)</sup>. Epigallocatechin gallate can also mimic insulin by increasing the tyrosine phosphorylation of both the insulin receptor and insulin receptor substrate-1, which

stimulates glucose uptake. In addition to the enhancement of insulin sensitivity, the possible factor underlying the attenuation of postprandial glucose concentrations in the GT group may be the inhibition of intestinal  $\alpha$ -amylase, sucrose and  $\alpha$ -glucosidase, which reduce the absorption of carbohydrates from the intestine<sup>(38)</sup>. Therefore, green tea catechins have some biological activities that can possibly provide anti-diabetic effects.

Another mechanism for improving glucose metabolism may be the improvement in oxidative stress including redox homeostasis<sup>(39)</sup>. It has been suggested that postprandial hyperglycaemia is positively correlated with the production of reactive oxygen species<sup>(10,40)</sup>. Indeed, ingestion of a high-carbohydrate or high-fat meal increases oxidative stress<sup>(11,22)</sup>. Particularly, intake of a high-fat meal results in greater postprandial oxidative stress than intake of a carbohydrate meal with the same energy content<sup>(11)</sup>. In the present study, serum d-ROM concentrations and plasma  $H_2O_2$  of oxidative stress markers increased significantly following the ingestion of high-fat meals. Alternatively, both acute and continuous (i.e. for 4 weeks) intakes of green tea following high-fat meals have no impact on the levels of postprandial oxidative stress markers. Despite no overall difference in the levels of oxidative stress markers between the GT and P groups, we found that an acute ingestion of catechin-rich green tea increases postprandial TRX concentrations. TRX plays an essential role in cellular function and protection by limiting oxidative stress directly via its antioxidant capacity<sup>(41)</sup>. Further support for this finding is provided by other studies that have found that the TRX system was up-regulated via the nuclear factor E2-related factor 2 signalling pathway by the consumption of green tea components including epigallocatechin-3-gallate<sup>(42,43)</sup>. Previous studies<sup>(13,44)</sup> have reported the relationship between glycaemic control disorders and oxidative stress. In diabetes mellitus, hyperglycaemia induces oxidative stress and contributes to the pathogenesis of diabetic vascular complications. Thus, elevated postprandial plasma TRX concentrations after an acute ingestion of catechin-rich green tea may have beneficial effects on oxidative stress status by mediating improved glucose metabolism in postmenopausal women.

Some investigators<sup>(16,32)</sup> have reported that green tea catechins attenuated resting oxidative stress markers. However, in the present study, we observed that both acute and continuous (i.e. for 4 weeks) ingestion of green tea catechins did not influence resting (i.e. fasting) oxidative stress and antioxidant capacity. It is unclear as to why the ingestion of green tea catechins in the present study had no effects on resting oxidative stress and antioxidant capacity. Similar to the results of oxidative stress and antioxidant capacity markers, resting glucose and insulin concentrations at baseline and at week 4 were not significantly different between the groups. This inconsistency may be explained by the fact that most studies investigated the effects of green tea catechins on resting oxidative stress and antioxidant capacity markers for a long period of time (ranging from 8 weeks to 3 months) compared with the present study<sup>(15,16,45)</sup>. Thus, long-term ingestion of green tea may be effective in improving resting glucose and oxidative stress status.

Oxidative stress and redox status are determined by the balance between oxidant and antioxidant levels<sup>(46)</sup>. It is important to note that oxidative stress markers and antioxidant capacity, which we measured in the blood, do not represent oxidative stress status in the cells. However, some studies<sup>(47,48)</sup> have reported that blood oxidative stress markers are associated with oxidative stress status in the tissues and cells. Moreover, there are several factors that influence and regulate redox status<sup>(49,50)</sup>. Therefore, we paid careful attention to several factors, including the nutritional contents of the beverage such as antioxidants (i.e. vitamin C and vitamin E) and the participants' physical activity (i.e. moderate to vigorous physical activity and step counts), medications, environmental factors and genetic factors. We can minimise these effects on blood redox status by setting up the exception criteria. Although we evaluated redox homeostasis, which was determined from serum TRX concentrations, the TRX system is a major antioxidant system that promotes the reduction of proteins by the cysteine thiol–disulphide exchange, which plays a vital role in maintaining the cellular redox balance<sup>(41,45)</sup>. Furthermore, to our knowledge, no information is available regarding the effect of green tea catechins on postprandial TRX concentrations in human subjects. Additional research is required to assess other redox parameters.

In conclusion, the present study demonstrates that an acute ingestion of catechin-rich green tea can decrease postprandial plasma glucose concentrations in postmenopausal women. This improvement may be explained by the fact that green tea ingestion elevated postprandial TRX concentrations, which suggests improved postprandial redox status, including TRX concentrations. However, continuous ingestion of catechin-rich green tea for 4 weeks showed no additive effects on the improvement of postprandial hyperglycaemia and oxidative stress in postmenopausal women.

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The authors' contributions are as follows: M. Takahashi designed the study, supervised the data collection, performed the blood and data analysis, and drafted the manuscript; M. M. conceived the study, obtained funding, recruited the participants, designed the study, assisted M. Takahashi with data collection and edited the manuscript; K. S. performed the venous blood collection and provided guidance and assistance to M. Takahashi during the study; S. B. and H.-K. K. assisted M. Takahashi with physiological and blood analysis; T. W., M. Takeshita and K. Y. designed the study, prepared the test beverages and provided guidance and assistance to M. Takahashi during the study; Y. M. designed the study, performed the blood catechin analysis in a double-blind manner, and provided guidance and assistance to M. Takahashi during the study. All authors read and approved the final manuscript.

Conflicts of interest: T. W., Y. M., M. Takeshita and K. Y. are employees of Kao Corporation. M. M. received a research grant from Kao Corporation. T. W., Y. M., M. Takeshita and



K. Y. were not involved in the interpretation of the results. The rest of the authors have no conflicts of interest to declare.

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