ELSEVIER

Nutrition 24 (2008) 562–568

NUTRITION

www.elsevier.com/locate/nut

Applied nutritional investigation

Association between soy and green tea (*Camellia sinensis*) diminishes hypercholesterolemia and increases total plasma antioxidant potential in dyslipidemic subjects

Márcia Bertipaglia de Santana, M.S.[a], Marcos Gontijo Mandarino, M.S.[b], Jefferson Rosa Cardoso, Ph.D.[c], Isaías Dichi, Ph.D.[d], Jane Bandeira Dichi, Ph.D.[d], Alissana Ester Iakmiu Camargo, M.S.[e], Bruno Alberto Fabris, M.S.[e], Ricardo José Rodrigues, M.D.[d], Elis Carolina Souza Fatel, M.S.[f], Suzana Lucy Nixdorf, Ph.D.[g], Andréa Name Colado Simão, M.S.[e], Rubens Cecchini, Ph.D.[h], and Décio Sabbatini Barbosa, Ph.D.[e,*]

[a] *Hospital Universitário de Londrina, Brazil*
[b] *Embrapa-Soja de Londrina, Brazil*
[c] *Departamento de Fisioterapia, Centro de Ciências da Saúde, Universidade Estadual de Londrina, Brazil*
[d] *Departamento de Clínica Médica, Centro de Ciências da Saúde, Universidade Estadual de Londrina, Brazil*
[e] *Departamento de Patologia, Análises Clínicas e Toxicológicas, Centro de Ciências da Saúde, Universidade Estadual de Londrina, Brazil*
[f] *Departamento de Nutrição, Faculdade Assis Gurgacz, Cascavel, Brazil*
[g] *Departamento de Química; Centro de Ciências Exatas, Universidade Estadual de Londrina; Brazil*
[h] *Departamento de Patologia; Centro de Ciências Biológicas, Universidade Estadual de Londrina, Brazil*

Manuscript received June 3, 2007; accepted February 6, 2008.

Abstract

Objective: To evaluate the hypolipemic and antioxidant effects of soy and green tea alone and/or in association in dyslipidemic subjects.

Methods: One hundred dyslipidemic individuals were allocated into four groups. The soy group ingested 50 g of soy (*kinako*) daily, and the green tea group ingested 3 g of green tea in 500 mL of water per day. A third group ingested 50 g of soy and 3 g of green tea daily, and the control group had a hypocholesterolemic diet. Evaluations were performed at baseline and after 45 and 90 d. Plasma levels of total cholesterol, high-density lipoprotein, and triacylglycerols were evaluated by automated methods. Low-density lipoprotein (LDL) cholesterol was calculated using the Friedewald equation. LDL was isolated by ultracentrifugation. Total plasma antioxidant capacity and plasma levels of total lipid hydroperoxides and those linked to LDL were evaluated by chemiluminescence. The results were expressed as median values and their 25th to 75th percentiles, with a 5% level of significance.

Results: No significant difference occurred in LDL, high-density lipoprotein cholesterol, and triacylglycerol levels across groups. However, a statistically significant difference in total cholesterol occurred within the soy/green tea group 45 and 90 d after intervention. No statistically significant difference occurred in plasma levels of lipid hydroperoxides or those linked to LDL in any of the groups studied. All the groups that used soy and/or green tea presented increased total plasma antioxidant potential.

Conclusion: Soy and green tea, alone or in combination, increased the total antioxidant potential of hypercholesterolemic patients, whereas only the combination decreased total cholesterol levels.

Keywords: Soy; Isoflavones; Green tea; Catechins; Antioxidants; Dyslipidemic human subjects

This study was supported by the Empresa Brasileira de Pesquisa Agropecuária (EMBRAPA-SOJA/Londrina), Kinasoy of Londrina, Midori Indústria de Chá Ltda., São Paulo, and the Clínica Cardiológica Centro de Doenças do Coração, Londrina, PR.

* Corresponding author. Tel.: +55-0-43-3371-2200; fax: +55-0-43-3371-2323.
E-mail address: sabatini@sercomtel.com.br (D. Sabbatini Barbosa).

0899-9007/08/$ – see front matter
doi:10.1016/j.nut.2008.02.007

Introduction

In the past 10 y, numerous researchers and the food industry have demonstrated growing interest in polyphenols. The main reason is the recognition of their antioxidant properties, their great abundance in nature, and their probable role in the prevention of several pathologies associated with oxidative stress, such as cancer and cardiovascular and neurodegenerative disorders. The beneficial effects of these substances depend on the quantity consumed and their bioavailability [1].

Several studies have considered that the hypocholesterolemic effect of vegetable proteins, in particular soy, is due to the high fecal excretion of steroids as a consequence of intestinal absorption reduction. A large fecal excretion of steroids could lead to increased biliary acid production from plasma cholesterol, thus reducing its own levels [2]. Moreover, soy protein amino acids and isoflavones appear to exert synergic action on improving lipid metabolism with the activation of receptors for low-density lipoprotein (LDL) [3].

From the chemical viewpoint, polyphenol compounds (such as isoflavones or flavanoids found in soy, tea, or wines) are the most promising natural antioxidants with regard to the inhibition of atherogenic modifications of LDL [4]. Even though the action mechanism of these polyphenols is not fully understood, the hypolipemic and protective actions against LDL oxidation are attributed to genistein and daidzein, in the case of soy, and to catechins, in the case of tea [5,6]. Considering the lack of consensus regarding these actions in the use of soy and green tea and, to our knowledge, the lack of studies that have made use of this association in humans to evaluate the lipid profile and analyze the total plasma antioxidant potential, the present study is justified.

Materials and methods

Casuistic

One hundred four hypercholesterolemic patients, 40 male and 64 female, were selected at a cardiology clinic in Londrina, Paraná, Brazil. Four patients, one male and three female, left during the development of the protocol for personal reasons. Therefore, 100 individuals, 39 male and 61 female, with a mean age of 52.2 ± 12.2 y, participated in the study.

The patients were sedentary and oriented to make no changes in their habitual lifestyle. This was monitored by means of a questionnaire that was filled out every time the patients returned to the clinic for blood collection. Furthermore, compliance was measured by this questionnaire and counting green tea and soy packages when patients returned for their clinical and nutritional evaluations. Body weight was measured to the nearest 0.1 kg by using an electronic scale, with individuals wearing minimal clothing; height was measured to the nearest 0.1 cm by using a stadiometer and was used to calculate body mass index (BMI; kilograms per square meter). The baseline BMIs (mean ± SD) for each group (green tea, soy, soy/green tea, and control) were 26.2 ± 3.3, 25.4 ± 3.2, 26.1 ± 3.3, and 27.1 ± 3.3 kg/m^2, respectively. Subjects in each comparison consumed similar dietary fat.

The inclusion criteria were patients >18 y old, with total cholesterol plasma levels ≥220 mg/dL. Individuals who presented with diabetes mellitus, hypothyroidism, or other causes of secondary dyslipidemia were excluded from the study. Women receiving hormone replacement therapy and patients who were using hypolipemic medication, isoflavones, soy, and/or green tea for ≥40 d before entry into the protocol were also excluded. No modification occurred in the treatment of patients receiving hypertensive medication.

All participants were informed regarding the procedures they would undergo and signed a term of free informed consent. This study and the term were approved by the ethics in human research committee of the Londrina University Hospital.

Study design: randomized controlled trial

Patients were randomized to one of four groups (green tea, soy, soy/green tea, and control) using sealed opaque envelopes indicating treatment allocation. Randomization envelopes were prepared at study inception and a random number sequence was obtained from a table of randomized numbers. After orientation by a nutritionist, participants used the products they were offered for 90 d. The soy group (n = 25) ingested 50 g/d of *kinako* (toasted ground soy bean, with 31.2 mg of isoflavones) throughout their daily meals: breakfast, lunch, and dinner. The green tea group (n = 25) ingested 500 mL of tea (dry leaves 3 g/vol, prepared by brewing for 10 min in 500 mL of water containing 145 mg of epigallocatechin-3-gallate [EGCG]) throughout the day. The soy/green tea group (n = 25) ingested the same as the soy and green tea groups. The control group (n = 25) received a standard hypocholesterolemic diet low in saturated fat and cholesterol contents and high in polyunsaturated fatty acids (PUFAs) [7]. From knowledge of the habitual intake of each individual, the control diet consisted of decreased red meat intake (beef and pork, a daily habit in Brazil), eggs, bacon, and replacing milk and butter with skimmed milk and margarine.

All groups were evaluated on three occasions by a blind assessor: at baseline and after 45 and 90 d. The soy was provided by the Brazilian Farming Research Company (EMBRAPA-SOJA, Londrina, Paraná, Brazil) and processed by the Kinasoy Company of Londrina. The green tea brand used was Yamamotoyama (Midori Indústria de Chá Ltda., São Paulo, Brazil) and donated by the manufacturer.

The batches and procedures of the products used by these groups were the same.

Analytical procedures

Extraction and separation of isoflavones for high-performance liquid chromatography

Separation and quantification of the isoflavones was performed using a Waters liquid chromatograph (model 2690, Watters & Associates, Milford, MA, USA) with an automatic sample injector [8]. The patients ingested the following concentrations of aglycone isoflavones in milligrams-percent (m/m) per 100 g of *kinako*: daidzein 23.2, glycitein 31.2, and genistein 8.0.

Determination of catechin EGCG by high-performance liquid chromatography in green tea samples

Separation and quantification of EGCG was performed using a Waters liquid chromatograph (model 2690) with an automatic sample injector [9]. The concentration of EGCG in 100 g of green tea leaves used by the patients was 4.84 g (m/m). The amount of EGCG present in the green tea ingested by the patients was 145 mg/500 mL.

Blood collection

Approximately 20 mL of blood was collected in vacuum tubes containing ethylene-diaminetetra-acetic acid or without a coagulant. Immediately after collection, the samples were chilled on ice. To obtain the plasma, the tubes were placed in a Fanem refrigerated centrifuge (Model 280 R, Guarulhos, São Paulo, Brazil) at 4°C and rotated at 3000 × *g*.

LDL separation

To obtain the LDL level, the following gradient was prepared: 0.5 mL of saline (density [d] = 1.182 g/mL), 3.0 mL of plasma (d = 1.090 g/mL), 1.0 mL of saline (d = 1.056 g/mL), 1.0 mL of saline (d = 1.045 g/mL), 1.5 mL of saline (d = 1.034 g/mL), 1.5 mL of saline (d = 1.024 g/mL), and 1.0 mL of saline (d = 1.019 g/mL). The pH of all these saline solutions was adjusted to 7.4 [10]. This gradient was centrifuged in a Sorvall ultracentrifuge (model Pro-80, Sorvall, Newtown, CT, USA), at 284 000 × *g* for 24 h at 4°C.

From the top of the tube, aliquots were removed in the following order: 1.0 mL, the aliquot considered to be very LDL; 6.0 mL, the aliquot considered to be LDL; and 3.5 mL, the aliquot considered to be infranatant.

Next, the LDL aliquot was dialyzed in 0.9% NaCl solution for 12 h to remove the excess KBr.

Plasma parameters

Biochemical quantifications were measured in a biochemical autoanalyzer (Dade AR, Newark, NJ, USA) using Dade Behring kits. Total cholesterol was quantified by the cholesterol oxidase technique. Whenever possible, LDL cholesterol was calculated by the Friedewald equation [11]. High-density lipoprotein (HDL) cholesterol was quantified by the selective precipitation method using a buffered phosphotungstate reagent. Triacylglycerols were analyzed by means of the bichromatic enzymatic technique, using lipase and glycerol dehydrogenase.

Total plasma antioxidant capacity

Total plasma antioxidant capacity (TRAP) was evaluated by chemiluminescence [12]. This methodology detects hydrosoluble antioxidants in the plasma. The 2,2 azobis generates peroxyl radicals rapidly, through an interaction centered on carbon and molecular oxygen, causing the oxidation of lipids and proteins in biomolecules [13]. These free radicals react with luminol, which acts as a signal amplifier, producing chemiluminescence. This reaction is inhibited by superoxide dismutase, catalase, and vitamin E analogs. The results were expressed as micromolar Trolox. This experiment was conducted in a Beckman β counter (model LS 6000, Beckman, Fullerton, CA, USA) in non-coincident counting mode for 30 min and with a response range of 300–620 nm.

Lipid hydroperoxides

Evaluation of the formation of lipid hydroperoxides was also realized by chemiluminescence [14]. The reaction medium was placed in 20-mL plastic flasks for scintillation. The medium consisted of 1750 μL of 30 mM phosphate buffer, pH 7.4, and 20 mM (v/v) KCl added to 250 μL of plasma or LDL and 20 μL of *tert*-butylhydroperoxide at a final concentration of 3 mM in 2.0 mL of reaction medium.

Chemiluminescence was measured in a Beckman β counter (model LS 6000) in non-coincident counting mode for 30 min and with a response range of 300–620 nm. The scintillation flasks were maintained in darkness and the reaction was realized in a dark environment to avoid phosphorescence of the flasks. This experiment was conducted at a temperature of 30°C and the results were expressed in counts per minute.

Statistical analysis

Initially, a descriptive analysis of the data was performed. Because the variables did not confirm to the assumptions of normality (lipid and antioxidant profiles), the data were presented as median and 25th to 75th quartiles. The percentage of change of outcome measurements was performed between baseline and final assessment (90 d). To compare the variables and percentage of change among and within groups, at three moments in time, analysis of variance for repeated measures was used. Mauchly's sphericity test was applied, and when it was violated, technical corrections were performed using the Greenhouse-Geisser test. When the F test was significant, complementary analysis was performed using Bonferroni's test for multiple comparisons. A statistical significance of 5% was adopted ($P \leq$

0.05). All analyses were performed with the intention to treat. SPSS 11.5 for Windows (SPSS, Inc., Chicago, IL, USA) was used for data analysis.

Results

Mean ages were 51.3 ± 13.4 y in the green tea group (9 men and 16 women), 52.7 ± 10.8 y in the soy group (11 men and 14 women), 57.0 ± 11.2 y in the soy/green tea group (8 men and 17 women), and 48.0 ± 12.4 y in the control group (11 men and 14 women).

The TRAP presented statistically significant differences within all groups and between the green tea and control groups. After the 90-d intervention, the tea group presented a significant difference compared with the control group and with percentage of change (17% of improvement in the green tea group versus 30.9% of worsening in the control group, $P < 0.0001$). After 45 and 90 d of treatment with green tea, there was a significant difference ($P = 0.007$) from baseline values. In relation to the soy group, significance was found when comparing baseline and 45-d interventions ($P < 0.001$) with 19% of change. In the soy/green tea group, significant differences were found between the baseline evaluation and the 45-d intervention ($P = 0.006$) and between the baseline evaluation and the 90-d intervention ($P = 0.007$) with 14% of change. With the exception of the control group, at the end of intervention, all groups presented an increase in antioxidant defenses. Statistically significant increases in TRAP occurred for the soy, green tea, and soy/green tea groups and a significant decrease in the control group (Table 1).

Regarding the pro-oxidant profile, no statistically significant differences occurred in plasma lipid hydroperoxide levels (23%, 45%, −14%, and 26% in each group, respectively) or in isolated LDL hydroperoxide levels (15%, −3%, −11.7%, and −16%) at any evaluation moment for all groups studied.

No statistically significant differences occurred between BMI and abdominal circumference when comparing the groups studied (data not shown).

Regarding the lipid profile, no statistically significant differences were observed in the concentrations of triacylglycerols (−12%, 13%, 36%, and 36%), HDL cholesterol (1.1%, 5%, 5%, and 0.6%) and LDL cholesterol (−6%, 6%, 3%, and −1.6%) among groups. However, total cholesterol presented a statistically significant reduction within the soy/green tea group ($P = 0.002$) between the baseline evaluation and after the 45-d intervention and 90 d ($P = 0.028$) with 6% of change (Table 2). The percentages of change in the other groups were −2%, 6.9%, and 3.7%.

Discussion

Diets based on polyphenols, substances renown for their antioxidant power, have been associated with a decrease in cardiovascular disorders [15]. Among soy-based products, *kinako* (toasted ground soy beans) possess a large concentration of isoflavones [16]. The consumption of approximately 50 mg of isoflavones per day is recommended for ideal hypocholesterolemic action [17]. In this study, the soy and soy/green tea groups ingested 31.2 mg of isoflavones per day. In the tea ingested by the patients, the daily concentration of EGCG was 145 mg/500 mL (3 g of leaves in 500 mL of water). In the groups under study (soy, green tea, soy/green tea), statistically significant increases in TRAP were observed. Similar results in relation to soy use were

Table 1
Medians and their quartiles (25th–75th) of TRAP, total LOOH, and LDL-LOOH in all groups studied

	Green tea group		Soy group		Soy/green tea group		Control group	
	n	Median (25–75)	*n*	Median (25–75)	*n*	Median (25–75)	*n*	Median (25–75)
TRAP								
Baseline	25	759 (636–895)	25	791 (751–854)	25	738 (599–785)	25	920 (838–1066)
45 d	25	928 (804–1073)†	25	1003 (933–1170)‡	25	930 (834–1000)§	25	767 (657–931)‖
90 d	25	958 (829–1076)*†	25	932 (818–1045)	25	937 (821–1048)§	25	728 (658–796)*‖
LOOH								
Baseline	25	21 900 (16 050–26 280)	25	21 932 (17 125–23 650)	25	17 450 (13 837–24 685)	25	14 000 (9600–14 750)
45 d	25	17 000 (10 850–24 500)	25	18 545 (13 727–23 635)	25	18 800 (15 470–21 700)	25	10 700 (8300–15 950)
90 d	25	18 500 (14 550–27 650)	25	18 800 (13 000–22 000)	25	17 500 (13 100–23 800)	25	13 000 (7800–17 250)
LDL-LOOH								
Baseline	25	9927 (8669–13 522)	25	11 371 (8357–17 549)	25	14 172 (8093–17 393)	25	4471 (2967–6984)
45 d	25	6466 (4230–9665)	25	9958 (5567–12 818)	25	11 594 (59 765–19 502)	25	4246 (1514–7193)
90 d	25	7284 (3297–12 287)	25	11 670 (5547–15 253)	25	15 124 (8487–25 330)	25	5453 (3516–10 968)

LDL-LOOH, low-density lipoprotein lipid hydroperoxides; LOOH, lipid hydroperoxides; TRAP, total plasma antioxidant capacity

* Statistically significant difference between green tea and control groups ($P < 0.0001$).

† Statistically significant difference within green tea group, baseline versus 45 d ($P = 0.007$) and baseline versus 90 d ($P = 0.007$).

‡ Statistically significant difference within soy group, baseline versus 45 d ($P = 0.001$).

§ Statistically significant difference within soy/green tea group, baseline versus 45 d ($P = 0.006$) and baseline versus 90 d ($P = 0.007$).

‖ Statistically significant difference within control group, baseline versus 45 d ($P = 0.02$) and baseline versus 90 d ($P = 0.001$).

Table 2
Medians and their quartiles (25th–75th) of total plasma cholesterol, HDL, LDL, and TG in the groups studied at each stage

	Green tea group		Soy group		Soy/green tea group		Control group	
	n	Median (25–75)	n	Median (25–75)	n	Median (25–75)	n	Median (25–75)
Total cholesterol								
Baseline	25	240 (228.5–274.5)	25	263 (247.5–292.5)	25	257 (244–290)	25	257 (244.5–276)
45 d	25	250 (228–269.5)	25	262 (240.5–309.5)	25	245 (224.5–261)*	25	250 (234–264)
90 d	25	264 (239–278.5)	25	259 (240–306.5)	25	246 (237.5–269)†	25	255 (232–279.7)
HDL								
Baseline	25	44 (35.5–54.5)	25	44 (32.75–53.0)	25	45.5 (37.75–53.50)	25	44 (38.5–53.5)
45 d	25	43 (34–53.5)	25	44.5 (33.50–59.25)	25	44 (39.25–58.0)	25	46 (38–54.5)
90 d	25	45 (34.5–56.5)	25	45 (38.25–58.75)	25	49.5 (44.5–56.0)	25	45 (35.7–51.0)
LDL								
Baseline	25	163 (143.50–202)	25	181 (160.5–215)	25	187 (167–196)	25	178 (148.5–196.5)
45 d	25	172 (156–181.75)	25	177 (161.2–199.75)	25	170.5 (156.75–192.25)	25	173 (153.5–189.5)
90 d	25	180 (166–206)	25	177.5 (154–205.75)	25	183 (162.25–193.25)	25	184.5 (146.5–203.7)
TG								
Baseline	25	132 (88–198)	25	177 (122.5–242)	25	170 (109.5–295.5)	25	147 (122.5–226.5)
45 d	25	149 (100.5–198)	25	176 (136–215)	25	141 (92.5–247.5)	25	159 (118–201)
90 d	25	144 (98–168.5)	25	174 (114–242.5)	25	128 (78–241.5)	25	127 (92.75–203.25)

HDL, high-density lipoprotein; LDL, low-density lipoprotein; TG, triacylglycerol
* Statistically significant difference within soy/green tea group, baseline versus 45 d ($P = 0.002$).
† Statistically significant difference within soy/green tea group, baseline versus 90 d ($P = 0.028$).

found by other investigators in a study involving women who ingested 40 g of soy-based products with 110 mg of isoflavones for 4 wk [18].

In a study involving healthy volunteers who ingested 300 mL (5.0 g of leaves) and 450 mL (7.5 g of leaves) of green tea, a significant increase in TRAP was also obtained [19]. Similarly, other researchers have observed an expresive improvement in TRAP when offering five cups of green tea per day over 4 wk to their patients [20]. According to the investigators, the increase in plasma antioxidant defenses verified by TRAP was important for the prevention of oxidation compounds present in the blood [20].

In the control group, in which a reduction in the ingestion of cholesterol and saturated fatty acids and an increase in PUFAs was planned, a statistically significant reduction in antioxidant defenses occurred, also verified by TRAP. This may be due to increased ingestion of PUFAs because the individuals of this group maintained their BMI after 90 d, revealing that no alteration in caloric ingestion occurred during the study.

Diets rich in monounsaturated fatty acids generate particles of LDL that are less susceptible to oxidation when compared with the LDL isolated from individuals who ingest a PUFA-rich diet [21]. Some studies have indicated a large concentration of oxidation products, such as conjugated dienes or plasma hydroperoxides, after the consumption of diets involving a large quantity of PUFAs compared with diets rich in monounsaturated fatty acids. The formation of hydroperoxides in PUFA-rich diets is directly proportional to the quantity of double bonds contained in the fatty acids [22].

In a study involving young pregnant women, a high level of isoprostane (a product of lipid peroxidation) excretion was detected. These patients ingested diets rich in PUFAs, specifically linoleic acid. Because these acids can serve as substrate for lipid peroxidation, PUFA-rich diets could contribute to increased oxidative stress, which is an imbalance between the production of pro-oxidant and antioxidant substances in the organism [23].

Although all groups that ingested polyphenols presented increased TRAP, no reduction in the formation of lipid hydroperoxides was observed. In contrast, other investigators found significant reductions in lipid hydroperoxide levels in women who ingested soy protein for 4 wk [17].

The present study also presented no statistically significant reduction in hydroperoxide levels in relation to isolated LDL. Given the hydrophilic characteristics of isoflavones, they probably present certain difficulties when incorporating into LDL particles [24]. Thus, only a slight interiorization of isoflavones occurs in these lipoproteins. This fact could have contributed to the absence of lower hydroperoxide levels in the isolated LDL of the patients.

After studying the inhibition of lipid peroxidation in vivo by polyphenol-rich beverages, such as green and black teas and red and white wines, researchers observed that these increased plasma antioxidant potential by different degrees in healthy individuals. The investigators concluded that the association of diverse polyphenol classes could act synergetically, leading to a significant increase in antioxidant defenses in vivo [25].

In a recent meta-analysis of 17 studies, it was concluded that the risk of myocardial infarct diminished 11% with the ingestion of three cups of green tea per day [26]. The individuals who made up part of the present study presented moderate hypercholesterolemia, with the me-

dian varying from 240 to 263 mg/dL. A significant reduction in total plasma cholesterol occurred only in the soy/green tea group after 45 and 90 d of intervention compared with the baseline value. However, when comparing among total cholesterol, LDL, HDL, and triacylglycerols in all groups, no statistically significant differences were observed.

Some researchers have stated that soy-based diets possess no significant hypocholesterolemic action in normocholesterolemic individuals. Moreover, they have reported that the hypocholesterolemic effect is directly proportional to cholesterol concentration before intervention [27–29]. In a study assessing three soy protein bars per day to normolipidemic individuals for 2 wk, no alterations in total cholesterol, LDL, HDL, and triacylglycerols were observed at the end of the protocol [30].

In this study, no significant difference occurred in the green tea group in relation to the lipid profile. In a large populational study, after adjustment by multivariable linear models, some investigators concluded that healthy Japanese workers showed a 2% reduction in plasma cholesterol after ingesting 10 cups of green tea per day. However, no alterations occurred in HDL cholesterol and triacylglycerol levels [6]. According to the investigators, despite the minimal decrease, a 1% reduction in total cholesterol means a 2–3% diminished risk of cardiovascular disease in Western countries and 5% for the Chinese. In contrast, other investigators have found no association between green tea consumption and improvement in lipid profile [6]. Corroborating these results, in 13 epidemiologic studies involving green tea ingestion to verify its hypocholesterolemic action, only four showed a significant reduction in lipids [31].

In this work, improvement in the lipid profile of patients was not observed in those who ingested soy or green tea alone but only in the soy/green tea group.

Considering the present data, it is possible that no significant improvement in plasma lipid levels was observed in the soy and green tea groups due to the moderate dyslipidemia of these patients. The association was shown to be more effective in this respect.

The results of this research demonstrate that soy flour and green tea, alone or in association, increased TRAP. When combined, these substances presented a synergy in relation to the hypocholesterolemic property.

In conclusion, the present study could not prove that the products used by the patients help avoid cardiovascular disorders; however, it contributed by providing a clearer understanding of how they might assist toward this end. If soy and green tea in isolation were shown to be important for increasing TRAP, the same was not verified in relation to the lipid profile, where the combined ingestion of soy and green tea appeared to provide more satisfactory results.

References

[1] Manach C, Scalbert A, Morand C, Rémésy C, Jiménez L. Polyphenols: food sources and bioavailability. Am J Clin Nutr 2004;79:727–47.

[2] Belleville J. Hypocholesterolemic effect of soy protein. Nutrition 2002;18:684–6.

[3] Anderson JW. Diet first, then medication for hypercholesterolemia (editorial). JAMA 2003;290:531–3.

[4] Kapiotis S, Hermann M, Held I, Seelos C, Ehringer H, Gmeiner BMK. Genistein, the dietary derived angiogenesis inhibitor, prevents LDL oxidation and protects endothelial cells from damage by atherogenic LDL. Arterioscler Thromb Biol 1997;17:2868–74.

[5] Clarkson TB. Soy, soy phytoestrogens and cardiovascular disease. J Nutr 2002;132(suppl 3):566S–9.

[6] Tokunaga S, White IR, Frost C, Tanaka K, Kono S, Tokudome S, et al. Green tea consumption and serum lipids and lipoproteins in a population of healthy workers in Japan. Ann Epidemiol 2002;12:157–65.

[7] Krauss RM, Eckel RH, Howard B, Appel LJ, Daniels SR, Deckelbaum RJ, et al. AHA dietary guidelines. Circulation 2000;102:2284–99.

[8] Berhow MA. Modern analytical techniques for flavonoid determination. In: Buslig BS, Manthey JA, editors. Flavonoids in the living cell. New York: Klusher Academic; 2002, p. 61–76.

[9] Lotito SB, Fraga CG. Catechins delay lipid oxidation and α-tocopherol and β-carotene depletion following ascorbate depletion in human plasma. Proc Soc Exp Biol Med 2000;225:32–8.

[10] Griffin BA, Caslake MJ, Yip B, Tait GW, Packard CJ, Sheppherd J. Rapid isolation of low density lipoprotein subfractions from plasma by density gradient ultracentrifugation. Atherosclerosis 1990;83:59–67.

[11] Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low density lipoprotein cholesterol in plasma without use of the preparative ultracentrifuge. Clin Chem 1972;18:499–502.

[12] Repetto M, Reides C, Carretero MLG, Costa M, Griemberg G, Llesuy S. Oxidative stress in blood of HIV infected patients. Clin Chim Acta 1996;255:107–17.

[13] Yokozawa T, Nakagawa T, Kitani K. Antioxidative activity of green tea polyphenol in cholesterol-fed rats. J Agric Food Chem 2000;50:3549–52.

[14] Flecha BG, Llesuy S, Boveris A. Hydroperoxide-initiated chemiluminescence: an assay for oxidative stress in biopsies of heart, liver and muscle. Free Radic Biol Med 1991;10:93–100.

[15] Vita J. Polyphenols and cardiovascular disease: effects on endothelial and platelet function. Am J Clin Nutr 2005;81(suppl):292S–7.

[16] Hendrich S, Murphy PA. Isoflavones: source and metabolism. In: Wildman REC, editor. Handbook of nutraceuticals and functional foods. Boca Raton, FL: CRC Press; 2001, p. 55–75.

[17] Chester EA. Soy for cardiovascular indications. Am J Health Syst Pharm 2001;58:663–6.

[18] Bazzoli DL, Hill S, DiSilvestro RA. Soy protein antioxidant actions in active, young adult women. Nutr Res 2002;22:807–15.

[19] Sung H, Chun S, Park H, Yang S, Min WK. In vivo antioxidant effect of green tea. Eur J Clin Nutr 2000;54:527–9.

[20] Prior R, Cao G. Antioxidant capacity and polyphenolic components of teas: implications for altering *in vivo* antioxidants status. Proc Soc Exp Biol Med 1999;220:255–61.

[21] Binkoski AE, Kris-Etherton PM, Wilson TA, Mountain ML, Nicolosi RJ. Balance of unsaturated fatty acids is important to a cholesterol-lowering diet: comparison of mid-oleic sunflower oil and olive oil on cardiovascular disease risk factors. Am Diet Ass 2005;105(7):1080–6.

[22] Diniz YS, Cicogna AC, Padovani CR, Santana LS, Faine LA, Novelli ELB. Diets Rich in saturated and polyunsaturated fatty acids: metabolic shifting and cardiac health. Nutrition 2004;20:230–4.

[23] Scholl TO, Leskiw M, Chen X, Sims M, Stein TP. Oxidative stress, diet and the etiology of preeclampsia. Am J Clin Nutr 2005;81:1390–6.

[24] Tikkanen MJ, Addercreutz H. Dietary soy-derived isoflavone phytoestrogens. Could they have a role in coronary heart disease prevention? Biochem Pharmacol 2000;60:1–5.
[25] Serafini M, Laranjinha JAN, Almeida LM, Maiani G. Inhibition of human LDL lipid peroxidation by phenol-rick beverages and their impact on plasma total antioxidant capacity in humans. J Nutr Biochem 2000;11:585–90.
[26] Vinson JA, Teufel K, Wu N. Green and black teas inhibit atherosclerosis by lipid, antioxidant, and fibrinolytic mechanisms. J Agric Food Chem 2004;52:3661–5.
[27] Nestel P. Role of soy protein in cholesterol-lowering. How good is it? Arterioscler Thromb Vasc Biol 2002;22:1743–4.
[28] Tikkanen MJ, Wähälä K, Ojala S, Vihma V, Adlercreutz H. Effect of soybean phytoestrogen intake on low density lipoprotein oxidation resistance. Proc Natl Acad Sci USA 1998;95:3106–10.
[29] Lichtenstein AH, Jalbert SM, Adlercreutz H, Goldin BR, Rasmussen H, Schaefer EJ, et al. Lipoprotein response to diets high in soy or animal protein with and without isoflavones in moderately hypercholesterolemic subjects. Arterioscler Thromb Vasc Biol 2002;22:1852–8.
[30] Park D, Huang T, Frishman W H. Phytoestrogens as cardioprotective agents. Cardiology 2005;13:13–7.
[31] Yang CS, Landau JM. Effects of tea consumption on nutrition and health. J Nutr 2000;130:2409–12.