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Habitual Intake of Guaraná and Metabolic Morbidities: An Epidemiological Study of an Elderly Amazonian Population

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The aim of the present study was to evaluate the associations of metabolic disorders and anthropometric and biochemical biomarkers of lipid, glucose and oxidative metabolism and the habitual ingestion of guaraná (*Paullinia cupana*, Mart. Var. sorbilis) by an elderly population residing in the Amazon Riverine region of the Maués municipality (Brazil). A case-controlled study was performed that included 637 elderly (≥ 60 years of age) patients classified as either those who habitually drank guaraná (GI, *n* = 421) or those who never drank guaraná (NG, *n* = 239) based upon their self-reported intake of guaraná. Indeed, the prevalence of various metabolic disorders was associated with guaraná ingestion. The prevalence of hypertension, obesity and metabolic syndrome in the GI group was lower than the prevalence found in the NG group. The NG group exhibited lower systolic and diastolic blood pressure values. The males in the GI group exhibited a lower waist circumference, on average, than the circumference found in the NG group, whereas the females in the GI group had lower cholesterol (total and LDL-c) levels than the control cohort. Additionally, a significant association was found between lower levels of advanced oxidative protein product (AOPP) and habitual guaraná consumption. The results constitute the first epidemiological study to suggest a potentially protective effect of habitual guaraná ingestion against metabolic disorders in elderly subjects.

Keywords: Paullinia cupana; guaraná; metabolic syndrome; oxidative metabolism; obesity; hypertension

INTRODUCTION

The plant guaraná (*Paullinia cupana*, Mart. var. sorbilis) originates in Brazil, is rich in methylxanthines such as caffeine, theobromine and theophylline, and contains tannins, saponins, catechins, epicatechins, proanthocyanidols, as well as trace concentrations of many other compounds (Belliardo *et al.*, 1985). A study of the guaraná transcriptome performed by Ângelo *et al.* (2008) revealed the presence of important secondary compounds in this plant, including transcript sequences related to flavonoid metabolism. These results suggested that guaraná exhibits similarities to *Camelia sinensis* (green and black tea), a plant that has been shown to have several interesting functional properties (Babu and Liu, 2008).

There have been medicinal beverages using extracts of roasted guaraná seeds available since the pre-Colombian era (Smith and Atroch, 2007). Previous studies in experimental models as well as *in vitro* assays have described several biological effects that guaraná shares with green tea, such as antioxidant activity (Mattei *et al.*, 1998; Basile *et al.*, 2005; Jimoh *et al.*, 2007), antimicrobial effects (Da Fonseca *et al.*, 1994; Pinheiro *et al.*, 1987; Yamaguti-Sasaki *et al.*, 2007) and anticarcinogenic and antitumoral properties (Fukumasu *et al.*, 2006; Leite *et al.*, 2010). Additional studies in animals and human volunteers have demonstrated that guaraná ingestion exhibits important biological effects, such as an improvement in cognitive performance (Espinola *et al.*, 1997; Kennedy *et al.*, 2004) and an antidepressive effect (Campos *et al.*, 2005; Otobone *et al.*, 2007). Similar effects have also been described for green tea consumption (Feng *et al.*, 2010; Niu *et al.*, 2009).

The functional properties of guaraná that are potentially the most similar to those of green tea are those properties related to metabolic disorders. Investigations have shown that guaraná positively affects lipid metabolism (Lima *et al.*, 2005), enhances weight loss (Boozer *et al.*, 2001; Opala *et al.*, 2006), and increases basal energy expenditure (Bérubé-Parent *et al.*, 2005). Therefore, these data suggest that guaraná potentially conveys an antiobesity effect. Furthermore, guaraná exhibits a cardioprotective effect due to opposition to platelet aggregation (Bydlowski *et al.*, 1988, 1991).

However, in contrast to green tea, in which many epidemiological studies have been performed that consistently describe effects on metabolic disorders and components of metabolic syndromes (Imai and

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Nakachi, 1995; Sasazuki *et al.*, 2000; Iso *et al.*, 2006; Kuriyama *et al.*, 2006; Basu *et al.*, 2010, 2011), most studies investigating the effects of guaraná have been performed in experimental models or in clinical approaches that used guaraná mixed with other bioactive compounds. In this manner, Boozer *et al.* (2001) investigated the associations among guaraná, *Mahuang*, and obesity, and Bérubé-Parent *et al.* (2005) studied the metabolic effects of guaraná, multi-vitamin supplements and green tea extracts. Other controlled investigations testing the effects of guaraná have been performed in studies over a short time period and with a relatively low number of subjects.

The investigation of guaraná consumption is important, in a manner similar to the investigation of other potentially beneficial foods such as green tea, soybeans and red wine, in larger population groups. Therefore, the objective of our study was to analyse the association between guaraná consumption and the prevalence of obesity, hypertension, type 2 diabetes and dyslipidemia in an elderly population living in the Riverine region of the Maués municipality in Brazil. Additionally, the study evaluated the effect of habitual guaraná ingestion on anthropometric and biochemical biomarkers of lipid, glycemic and oxidative metabolism.

MATERIALS AND METHODS

Study design, population characteristics, and sample selection. There were a total of 637 elderly (≥60 years of age) patients included in the present case-controlled study. The elderly patients were classified into two groups based on self-reported data: those who habitually ingested guaraná and those who never ingested guaraná. At the time of data collection, Maués had 45284 inhabitants, of which 2939 (6.4%) were elderly. Therefore, the samples analysed represent 22.4% of the elderly population. This sample population was based upon a previous investigation performed by our research group that included 1808 elderly subjects (Ribeiro *et al.*, 2010).

The study participants comprised elderly volunteers who could be easily accessed by researchers; about 50% of the population lived in far-flung coastal communities located among the rivers and tributaries of the Amazon forest. The study age criteria of ≥60 years was based on the World Health Organization (WHO, 1998) definition of an 'elderly' population in a developing country. The baseline general characteristics of the subjects studied here are described in Table 1. Aside from the higher number of males in the GI group compared with females, the other socioeconomic and cultural variables were similar between the groups.

This study was performed in an elderly population because elderly people tend to have a stable dietary pattern and are less susceptible to changes arising from the increased global use of nutritional foods compared with young adults. Additionally, epidemiological studies have demonstrated that the metabolic diseases investigated here are most prevalent in elderly adults (Cigolle *et al.*, 2009).

Region of study and history of guaraná consumption. The study was performed in Maués because this location is historically important for guaraná production. Maués is located in the geographical middle of the Amazon region; it was founded in 1798 by the Portuguese and became a municipality in 1896. The prominent region of Maués is located on the right bank of the River Maués-Açu (Table 2).

The primary agricultural product of Maués is guaraná (*Paullinia cupana*). Evidence has suggested that the native Sateré-Maués people, who live in a native indigenous reserve localized in Maués, were the first to farm guaraná. However, as guaraná consumption increased rapidly among European colonizers, the beverage was incorporated into the traditional culture of the mixed population that arose from the interaction between the settlers and the indigenous peoples of

Table 1. Baseline characteristics of elderly Riverine inhabitants who habitually ingest guaraná (GI) and those who never ingest guaraná (NG), Maués, Amazonas-Brazil

Variable		Group		*p*
		GI	NG	
Sample number (*n*, 637)		421	239	
Sex	Males	219	86	<0.0001
	Females	202	153	
Age (years, mean ± SD)		72.87 ± 8.13	71.93 ± 7.84	0.149
Place of birth	Maués	304 (73.6)	156 (69.6)	0.384
Education				
	Illiterate	287 (68.8)	158 (68.1)	0.923
	<4 years	82 (19.7)	46 (19.8)	
	4–8 years	22 (5.3)	15 (6.5)	
	>8 years	26 (6.2)	13 (5.6)	
Functional status	Retired	360 (86.7)	192 (82.1)	0.135
Occupation	Subsistence farming[a]	396 (94.5)	234 (97.9)	0.203
Personal income		225 (55.6)	116 (51.3)	0.307

n = amostral sample.

[a]Agricultural and fishing activities; comparison between categorical variables were performed by Chi-square statistical test.

Table 2. Maués, Amazonas-Brazil baseline characteristics

Indicator	
Geographic location	Latitude: 3°38′36.1″S Longitude: 57°71 ′86.1″W
Area	39988 km^2
Population (2009)	45284
Transport	Access: boat and air transport
Distance from Manaus	356 km (train transport)
Population distribution (2009)	Urban Riverine region: 21094 Rural Riverine: 24190 Total: 45284 Elderly population: 2939 (6.4%)
Riverine population geographic distribution	175 communities distributed in several river deltas
Demographic density	1 hab/km^2
Life expectancy at birth[a]	68.3 years

[a]Brazilian Governmental Census 2000.

the Amazon (Smith and Atroch, 2007). This general population is known as the Riverine or Caboclos population. The origins of the Caboclos cultural group can be traced back approximately 300 years. In the Maués region Caboclos are also referred to as Ribeirinhos or Riverine people (river-side dwellers).

The Riverine people that live in Maués have developed several different traditional methods of guaraná production that have been described by Smith and Atroch (2007); generally, guaraná is produced on small and large farms, either as a monocrop or alongside other crops, and is harvested by hand in the dry season. If the entire fruit bunch is ripe, it is either snipped off with scissors or small pruning shears, or broken off manually and placed in a basket and carried back to the home. Before roasting the seeds, the red skin must be removed, and so the fruits are skinned by hand, left to soak in water, or simply stored for several days until the skin softens. On small farms in the Maués watershed the guaraná seeds are roasted on a griddle, usually made of clay, that reduces the chance of burning.

To prepare the traditional guaraná beverage, the hard cylinder containing the seeds is grated with the bony tongue of the pirarucu (*Arapaima gigas*), one of the largest fish in the Amazon. The powder is collected either on a piece of paper or allowed to fall directly into a calabash gourd containing water. Sugar is then added to the mixture of guaraná powder and water, and the whole concoction is consumed, usually soon after waking, while the consumer is still in the fasting state. The ingestion of guaraná more than once a day does occur, but it appears to depend on an individual's level of energy expenditure. Repeated intake of guaraná throughout the day may be related to extreme environmental conditions, such as high temperature and humidity, common conditions in the Amazon rainforest region that increase the feeling of fatigue. We used this information as background in a brief questionnaire about sociocultural, economics, health and lifestyle variables. In the group who reported never drinking guaraná, the primary motivations were dislike of the taste and the experience of tachycardia as a consequence of the high caffeine concentration. Elderly people who had previously been advised not to drink guaraná for medical reasons were excluded from this study. The elderly people who reported drinking guaraná noted that they had begun ingesting the drink at a young age, often as children. Moreover, the subjects who consumed guaraná reportedly ingested this plant at least two or more times a week.

Metabolic disorder diagnosis. Initially, the study evaluated the associations between guaraná ingestion and the following metabolic disorders: obesity, hypertension, type 2 diabetes, and metabolic syndrome. The subjects were classified into the following groups based on their body mass index (BMI): obese, BMI > 30 kg/m^2; overweight, BMI ≥ 25 kg/m^2, and <30 kg/m^2; control group (non-overweight), BMI < 25 kg/m^2. Hypertension was considered to be present when the subject had a systolic blood pressure (SBP) of >140 mmHg and/or a diastolic blood (DBP) pressure of >90 mmHg, when measured on a minimum of two occasions separated by a month, or when antihypertensive drugs were used. Subjects with severe hypertension, i.e. SBP ≥ 160 mmHg and/or DBP ≥ 100 mmHg, were also included. Diabetes mellitus (type 2) was considered to be present when two independent measures demonstrated that the subject had glucose levels above 126 mg/dL or if the subject was using glucose-lowering drugs. The metabolic syndrome was diagnosed when the participant met three or more of the following criteria: (1) high blood pressure: blood pressure ≥ 130/85 mmHg or under treatment for hypertension; (2) hypertriglyceridemia: fasting plasma triglycerides ≥ 150 mg/dL; (3) low HDL: fasting HDL cholesterol < 40 mg/dL in men, <50 mg/dL in women; (4) hyperglycemia: fasting glucose level of ≥110 mg/dL or under treatment for diabetes; (5) central obesity: waist circumference >88 or >102 cm in women and men, respectively (Grundy, 2005).

Also elderly patients without metabolic disorders or other morbidities (healthy elderly) were identified.

Biochemical and anthropometric data collection. Biochemical analyses were performed on blood samples collected from subjects after an overnight (>12 h) fast. Peripheral blood samples were collected by venipuncture using Vacutainer® (BD Diagnostics, Plymouth, UK) tubes containing heparin and EDTA. The levels of the following blood components were assayed: glucose, total cholesterol, HDL-c, LDL-c and triglycerides (TG). Total cholesterol, HDL-c, TG and glucose levels were determined using an enzymatic colorimetric method with the following commercial kits: total cholesterol, Cod-Ana Labtest®; HDL-c, precipitant Labtest®; TG, Gpo-Ana; Glucose, PAP Labtest®. The LDL-c level was calculated according to the Friedewald equation: (LDL-c) = (TG) − (HDL-c + TG/5) (Tonks, 1972; Freidewald *et al.*, 1972).

The levels of biomarkers of oxidative metabolism were analysed; the total polyphenol content was spectrophotometrically determined in plasma by measuring the absorbance at 750 nm (the Folin-Ciocalteau method) and using gallic acid as a standard, as described by Chandra and De Mejia Gonzalez (2004). The total phenol concentration of the plasma samples was determined after a procedure of acid extraction/hydrolysis and protein precipitation with 0.75 M metaphosphoric acid (MPA). For hydrolysis of the conjugated forms of polyphenols, hydrochloric acid was added to the sample, followed by sodium hydroxide in methanol, which breaks the polyphenol–lipid links and provides a first extraction of polyphenols. MPA was used in this procedure to remove the plasma proteins. The final extraction of polyphenols was performed by adding a 1:1 (v/v) solution of acetone:water. The results were expressed as the gallic acid equivalent (GAE) in mg/L, and thiol groups were determined as described by Ellman (1959). Lipid peroxidation was quantified by measuring the formation of thiobarbituric acid reactive substances (TBARS) (Ohkawa *et al.*, 1979). Total blood SOD (EC 1.15.1.1) activity was measured spectrophotometrically according to Boveris and Cadenas (1997). One unit of activity is defined as the amount of enzyme required to inhibit the rate of epinephrine auto-oxidation by 50%. Catalase activity (EC 1.11.1.6.) was determined according to Aebi (1984). One unit of catalase activity was defined as the activity required to degrade 1 μmol of hydrogen peroxide in 60 s. Protein carbonyls were measured according to a method described previously (Morabito *et al.*, 2004). The results were expressed as nanomoles of carbonyl groups per mg protein. Advanced oxidized protein products (AOPP) were measured using a Cobas Mira Plus clinical chemistry analyser and using the technique described by Selmeci *et al.* (2005). Nitric oxide was evaluated indirectly by plasma nitrate and nitrite quantification using the Griess method (adapted to Cobas Mira automated analyser by Pereira *et al.* (2010)).

The anthropometric variables investigated included height (measured in meters; without shoes) and weight (measured in kilograms; with heavy clothing removed and 1 kg deducted for remaining garments). Body mass index was calculated as weight in kilograms divided by the square of the height in meters. The waist circumference on standing subjects was measured with a soft tape midway between the lowest rib and the iliac crest. Two blood pressure recordings were obtained from the right arm of patients in a sitting position after 30 min of rest; measurements were taken in 5 min intervals, and the mean values were calculated.

Ethics. This study was approved by the Ethical Committee of the Universidade do Estado do Amazonas. Since the vast majority of the elderly included in this study were illiterate, oral consent or fingerprint in Term was obtained to indicate their voluntary participation in the study after the researchers read the consent form to the patients.

Statistical analysis. Statistical analysis was performed using the SPSS/PC statistical package, version 17.0 (SPSS, Inc., IL). The difference in the prevalence of metabolic diseases between the elderly subjects who habitually ingested guaraná and the elderly subjects who never consumed guaraná was compared using the Chi-square test. A multivariate analysis was performed using logistic regression (the backward Wald method) to evaluate age, gender, smoking and previous cardiovascular disease as possible intervening variables. The biomarker comparison was performed using the Student's *t*-test analysis and considered males and females separately due to the potential biological differences between sexes, such as waist circumference. The variables that presented statistical significance were tested using multivariate analysis in order to observe whether the associations were independent of age, smoking habits, medicine consumption, previous cardiovascular disease and risk factors. The odds ratio (OR) and confidence interval at 95% (CI95%) were calculated for categorical variables associated with guaraná consumption. All p values were two-tailed, and $p < 0.05$ was considered statistically significant.

RESULTS

The prevalence of metabolic disorders in the GI and NG group was analysed, and the results are shown in Table 3. The prevalence of metabolic disorders was negatively associated with guaraná ingestion. The group that consumed guaraná showed a lower prevalence of hypertension, obesity and metabolic syndrome than the NG group. However, the prevalence of type 2 diabetes was identical between the GI and NG groups. Multivariate analysis showed that these results were independent of age and sex. For hypertension in the GI group, the odds ratio (corrected for sex and age) was 0.699 (95% CI = 0.500 – 0.960). For obesity the odds ratio was 0.661 (95% CI = 0.456 – 0.958) and for metabolic syndrome it was 0.856 (95% CI = 0.726 – 1.000). These results indicate that there is a protective effect associated with habitual guaraná ingestion.

Three additional analyses related to health conditions among the elderly were performed. As shown in Table 2 the percent of subjects that had self-reported daily consumption of medicine was higher in the NG group than in the GI group. However, the incidence of hospitalization over the past year was similar between the two groups. The percent of subjects with no history of metabolic disorders or other morbidities was 19.9% ($n = 127$) and was not significantly different between the GI and NG groups ($p = 0.765$).

There were a total of 42 patients in this study who had self-reported histories of stroke or myocardial infarction; there was a significantly higher prevalence of these seniors in the group who did not drink guaraná ($n = 22$, 9%) than in the habitual consumption group ($n = 20$, 5%) ($p = 0.028$). These results were independent of sex and age.

Another important variable in the results described here is smoking; 12% ($n = 76$) of the subjects were

Table 3. Comparison of the prevalence of metabolic disorders between elderly Riverine inhabitants who habitually ingest guaraná (GI) and those who never ingest guaraná (NG)

Disorder	Group		p
	GI	NG	
	% (*n*)	% (*n*)	
Hypertension	43 (174)	53 (123)	0.018
Type 2 diabetes	11 (44)	15 (35)	0.186
Obesity	32 (131)	47 (108)	<0.001
Metabolic syndrome	21 (70)	32 (56)	0.007
Daily medicine ingestion	44 (185)	55 (129)	0.020
Hospitalization in the past year	13 (54)	15 (35)	0.562

n = amostral sample. Statistical comparison was performed using a Chi-square test.

smokers, and the frequency of smoking was similar between the GI and NG groups ($p = 0.509$).

The biochemical and anthropometric variables for the male and female patients were analysed separately due to biological characteristics such as abdominal circumference that differ between sexes. This analysis also excluded subjects with a history of type 2 diabetes mellitus, whose self-reported use of medicine could influence the variables investigated; the results obtained are shown in Table 4. The males in the GI group exhibited a significantly lower waist circumference, systolic blood pressure and diastolic blood pressure when compared with the males in the NG group and multivariate analysis showed that these factors were significantly associated with guaraná ingestion independent of age, smoking habit, diabetes and previous cardiovascular disease. However, the biochemical variables investigated were similar between the two groups, aside from the AOPP level, that was found to be lower in the GI group.

Females in the GI group exhibited lower systolic and diastolic blood pressure than the females in the NG group. In contrast to the results obtained in male subjects, waist circumference was found to be similar between the two groups. However, habitual ingestion of guaraná was associated with differences in some biochemical parameters. In females, total cholesterol, LDL-cholesterol and AOPP levels were significantly lower in the GI group compared with the NG group.

An additional analysis was performed using a level of 25 μmol/L AOPP between the GI and NG groups as a cut-off value in order to delineate the first quartile of the sample population. The frequency of subjects with lower AOPP values was significantly reduced in the GI group ($n = 117$, 38%) compared with the NG group ($n = 41$, 25%) ($p = 0.004$). The odds ratio for higher AOPP values in the NG group (after correction for sex, age, previous cardiovascular diseases and smoking) was OR = 1.709 (95% CI = 1.133–2.578).

DISCUSSION

This is the first epidemiological study to investigate the association between the prevalence of metabolic disease and habitual guaraná ingestion in an elderly Amazon rainforest population residing in a region of Brazil known for its guaraná production (Maués). In general, the results suggest that guaraná consumption, most likely due to the bioactive compounds present in the beverage, potentially conveys a protective effect against the metabolic disorders investigated here. The association between guaraná ingestion and a lower risk or obesity, hypertension and metabolic syndrome is important because these morbidities are related to cardiovascular disease risk. Some of the differences in the biochemical and anthropometric parameters that were observed in this study corroborate the possible protective effect of guaraná.

Our findings on the impact of habitual guaraná ingestion on the prevalence of metabolic disorders are in concordance with previous results obtained from experimental models and clinical investigations that used guaraná as a supplement. The antiobesity effects of guaraná have been described by Boozer *et al.* (2001), Bérubé-Parent *et al.* (2005) and Opala *et al.* (2006).

In addition to finding a lower prevalence of obesity in the elderly subjects who habitually ingested guaraná, an association was found between the consumption of guaraná and waist circumference (WC) in men. These data potentially have an epidemiological impact as waist circumference is considered to be a measure of abdominal obesity correlated with cardiovascular risk, particularly in men. Population studies have described positive associations between this trait and higher mortality, independent of BMI. A recent investigation by Jacobs *et al.* (2010) examined the association between WC and mortality among 48500 men and 56343 women aged 50 or more in the Cancer Prevention Study II

Table 4. Comparison of biochemical, anthropometric and physiological variables between elderly Riverine inhabitants who habitually ingest guaraná (GI) and those who never ingest guaraná (NG)

Variable	Male		Female	
	GI	NG	GI	NG
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
BMI (kg/m^2)	25.1 ± 5.6	25.1 ± 4.4^{ns}	26.0 ± 4.5	25.0 ± 4.1^{ns}
Waist circumference (cm)	87.3 ± 14.4	92.7 ± 11.3^{b}	86.7 ± 14.2	87.6 ± 16.4^{ns}
SBP (mmHg)	130.2 ± 20.4	137.3 ± 21.9^{b}	129.4 ± 18.6	134.1 ± 22.4^{a}
DBP (mmHg)	73.9 ± 11.1	76.9 ± 10.8^{a}	74.5 ± 9.9^{ns}	75.3 ± 10.4^{ns}
Glucose (mg/dL)	107.4 ± 16.5	110.1 ± 18.3^{ns}	118.9 ± 37.1	115.5 ± 31.1
Cholesterol (mg/dL)	193.6 ± 191.9	191.9 ± 41.3^{ns}	207.6 ± 48.3	227.8 ± 60.9^{b}
LDL-c (mg/dL)	144.3 ± 49.2	156.2 ± 59.3^{ns}	140.5 ± 45.0	155.9 ± 57.2^{a}
HDL-c (mg/dL)	37.9 ± 15.7	38.4 ± 11.2^{ns}	37.9 ± 15.7	38.4 ± 11.2^{ns}
Triglycerides (mg/dL)	178.9 ± 89.9	168.4 ± 85.4^{ns}	178.9 ± 89.9	168.4 ± 85.4^{ns}
Total polyphenols (mg/dL)	2.58 ± 0.67	2.60 ± 0.51^{ns}	2.56 ± 0.54	2.43 ± 0.70^{ns}
TBARS (nmol/mL erit)	21.8 ± 9.6	21.5 ± 10.2^{ns}	22.0 ± 9.9	21.3 ± 8.3^{ns}
Carbonyl proteins (mg/dL)	0.173 ± 0.09	0.183 ± 0.09^{ns}	0.183 ± 0.09	0.189 ± 0.08^{ns}
AOPP (mmol/L)	36.3 ± 30.9	44.3 ± 35.2^{a}	34.6 ± 19.6	41.1 ± 28.1^{a}
Nitric oxide (mmol/L)	30.5 ± 24.1	34.9 ± 26.8^{ns}	34.7 ± 28.5	34.5 ± 21.1^{ns}

SD, standard deviation; SBP, systolic blood pressure; DBP, diastolic blood pressure; AOPP, advanced oxidation protein products. Mean comparison between GI and NG groups were performed by statistical Student's *t*-test. NS, not significant; $^{a}p < 0.05$; $^{b}p < 0.01$

Nutrition Cohort. The authors observed that, after adjustment for BMI and other risk factors, very high WC values were associated with an approximately two-fold higher risk of mortality in men and women. Thus, the association between lower waist circumferences and ingestion of guaraná described here appears to be relevant. The catechins, caffeine, and other xanthines present in guaraná likely contribute to these results; however, the exact mechanism of action remains to be elucidated.

As noted previously, guaraná contains some bioactive compounds that are similar to those found in green tea (Belliardo *et al.*, 1985; Ângelo *et al.*, 2008) that have been well studied. Tea polyphenols have been shown to exhibit antioxidative, antithrombogenic, antiinflammatory, hypotensive, hypocholesterolemic, antihypertensive and antiobesogenic effects (Yung *et al.*, 2008).

An association was also observed between hypertension and metabolic syndrome and guaraná ingestion. However, the effect of guaraná on hypertension and metabolic syndrome in humans is poorly understood. On the other hand, a growing body of evidence indicates that there is a potential role for green tea, or its ingredient bioactive polyphenol epigallocatechin gallate (EGCG), in significantly ameliorating features of metabolic syndrome and subsequent risks for type 2 diabetes mellitus and cardiovascular disease. The results from these studies demonstrate the beneficial effects of green tea or green tea extracts rich in EGCG on weight management, glucose control and cardiovascular risk factors (Thielecke and Boschmann, 2009).

Certainly, the most unexpected result reported here, as guaraná contains a high concentration of caffeine, was the association between guaraná intake and the low prevalence of hypertension. The effect of caffeine on blood pressure has been examined for decades, with variable results that depend on factors such as the population under study. The relationship between foods rich in caffeine and hypertension is based on the extensive use of caffeinated drinks, particularly coffee, in modern societies. However, epidemiological studies have not found a consistent relationship between dietary caffeine intake and the incidence of hypertension (Smith *et al.*, 2003). An explanation for the lack of such a relationship is that regular caffeine consumption is thought to lead to complete tolerance of its effects on blood pressure (Myers and Reeves, 1991).

Therefore, a question that emerges from our results concerning blood pressure and hypertension and guaraná ingestion is whether these results are related to the development of tolerance to caffeine or whether the association is due to the existence of chemical interactions between that compound with other chemicals present in guaraná, including catechins. Evidence from robust longitudinal studies such as the Framingham Heart Study suggests that there is a significant negative association between the consumption of caffeinated coffee and cardiovascular events, i.e. lower risk of cardiovascular mortality heart valve disease development or progression in older Framingham subjects without moderate and severe hypertension (Greenberg *et al.*, 2008). A recent study (Chen *et al.*, 2010) examined the relationship between sugar- sweetened beverages, including coffee, and blood pressure in a prospective analysis that included 810 subjects. The authors observed that reduction of sugar in such beverages caused a reduction in blood pressure. The authors noticed no association between caffeine intake and blood pressure.

It is clear that hypertension, obesity and metabolic syndrome are common pathophysiological aspects that are directly impacted by nutritional status. Excessive energy intake and obesity are major causes of hypertension. Obesity is associated with increased activity of the rennin–angiotensin–aldosterone pathway and the sympathetic nervous system, as well as with mineralocorticoid activity, insulin resistance, salt-sensitive hypertension, excess salt intake and reduced kidney function (Savica *et al.*, 2010).

Recently, Namkung *et al.* (2010) found that tannic acid and the related gallotannins present in green tea and red wine exhibited an inhibitory effect on Ca^{2+}-activated Cl^- channels (CACCs). The authors concluded that gallotannins are potent CACCs inhibitors whose biological activity provides a potential molecular basis for the cardioprotective and antisecretory benefits of red wine and green tea. Therefore, there is a need for the performance of complementary studies in order to evaluate whether the tannins and catechins present in guaraná have similar effects on blood pressure.

In addition to examining the prevalence of metabolic disorders, we also investigated whether habitual intake of guaraná exhibited an impact on components of oxidative metabolism. It was decided to investigate this as previous evidence has shown that polyphenols (such as catechins) present in beverages such as green tea and guaraná exhibit antioxidant and antiinflammatory properties (Mattei *et al.*, 1998; Basile *et al.*, 2005; Babu and Liu, 2008). This study describes an important result regarding the association between guaraná ingestion and AOPP levels.

Protein oxidation and glycosylation are post-translational modifications implicated in the pathological development of many age-related diseases (Zwart *et al.*, 2009). Changes in AOPP levels are connected to poor glycemic control, chronic disease, dyslipidemia and diabetic complications, particularly nephropathy (Cakatay, 2005).

A significant association was found between higher AOPP levels (≥25 μmol/L) and lack of guaraná consumption. It is important to note that our choice of the cut-off value (≥25 μmol/L) was based on a percentile distribution of AOPP values in our sample population and represented the first quartile. However, in the study by Selmeci *et al.* (2005) that established reference values for AOPP in a student population aged 18–33 years this cut-off point lies approximately in the 3rd quartile. We believe that the discrepancy could be caused by age because our sample population was composed of elderly volunteers. A previous study performed by Pandey *et al.* (2010) reported that oxidative alterations in biomarkers of plasma protein oxidation such as protein carbonyls (PCO), plasma total thiol groups (ThG) and AOPP are age-dependent; with increased age, PCO and AOPP levels increase and ThG levels decrease. In an additional investigation performed by Pandey *et al.* (2010), healthy control subjects with a mean age of 58 ± 7 years exhibited an AOPP concentration of 63.64 ± 25.23 μmol/L, whereas the mean AOPP level in diabetic patients was significantly higher (89.51 ± 36.46 μmol/L). It is of interest to point out that the mean AOPP value in our sample population was lower than that described by Pandey and collaborators regarding the healthy control group. A greater number of habitual guaraná drinkers exhibited low levels of AOPP, suggesting a protective effect of guaraná that is related to protein oxidative damage.

Studies investigating the association between AOPP levels and ingestion of phytotherapeutic beverages such as green tea have also been performed. Nakagawa *et al.* (2002) investigated the potential effects of green tea extract, a green tea tannin mixture and its components, on protein damage induced by 2,2′-azobis(2-amidinopropane) dihydrochloride (a free radical generator) and glucose *in-vitro*. The authors reported that the green tea extract effectively conferred protection against protein damage and that this protection was most likely due to the presence of a tannin compound; these results suggest that green tea could be useful in the control of protein oxidation- and glycosylation-associated diseases.

Due to the fact that nitric oxide (NO) participates in such highly active metabolic and regulatory processes as hemostasis, fibrinolysis in platelets, vascular tone modulation and blood pressure homeostasis, plasma nitrite, an indirect measure of NO release, was evaluated in our sample population. Although the nitrite levels described here are similar to those in other adult populations (Ghasemi *et al.*, 2010), no association was found between guaraná ingestion and plasma levels of nitrite or other oxidative biomarkers.

Therefore, although previous studies have described antioxidant properties of guaraná based on results obtained from in vitro assays and other experiments (Mattei *et al.*, 1998; Basile *et al.*, 2005; Jimoh *et al.*, 2007) the effect of guaraná on such markers of oxidative metabolism as AOPP levels in humans has, to our knowledge, never before been described.

It is important to comment on the methodological limitations of this study. Because this investigation used a cross-sectional design, it is not possible to determine whether the associations found represent cause-and-effect relationships. As we selected the case–control subjects of the population and, as in the previous investigation in which 1808 subjects were analysed, a higher prevalence of guaraná consumption was found in males compared with females, thus it was decided to maintain this proportion in the investigation described here.

There is no previous reference to explain the higher consumption of guaraná by males compared with females. However, guaraná is traditionally consumed in the region for its antifatigue and aphrodisiac properties. Therefore, the labor-intensive fishing and agricultural work commonly performed by males is potentially the culturally associated reason behind the higher consumption of guaraná. Thus, in our study we opted to maintain this proportion in the case–control groups and to evaluate the possible influence of gender using statistical tools such as multivariate analyses.

Furthermore, other variables exist that were not investigated in this population, such as physical activity and genetic factors, and these variables potentially influence the results to some extent. However, the results described here are intriguing, and are in concordance with previous studies in various experimental models, and moreover are similar to results obtained with other herbal beverages such as green tea.

In this context, we believe that, despite the methodological limitations, the results described in this study suggest that habitual guaraná ingestion contributes positively to the prevention of various metabolic disorders in the elderly.

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Conflict of Interest

The authors have declared that there is no conflict of interest.

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