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Antioxidant Potential of Rat Plasma by Administration of Freeze-Dried Jaboticaba Peel (*Myrciaria jaboticaba* Vell Berg)

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ABSTRACT: The effect of the intake of freeze-dried jaboticaba peel powder on the antioxidant potential of rats' blood plasma was investigated in two experiments. In the first, 35 male rats, divided into 5 groups, received 7 mg of anthocyanins/100 g of body weight, by gavage. The blood was withdrawn 0, 30, 60, and 120 min after gavage. The antioxidant capacity was evaluated in plasma by ORAC and TEAC. There was no significant difference in the plasmatic antioxidant potential among the groups. In the second experiment, 40 male rats were divided into 4 groups that consumed, respectively, 0, 1, 2, and 4% of freeze-dried jaboticaba peel powder added to their diet. An increase in the plasmatic antioxidant potential was observed for groups that received 1 and 2% of jaboticaba peel powder (1.7 times by TEAC method and 1.3 times by ORAC); however, the group that received 4% of the powder did not show antioxidant effects according to the methods used.

KEYWORDS: anthocyanins, antioxidant, jaboticaba, *Myrciaria jaboticaba* Vell Berg, ORAC, TEAC

INTRODUCTION

Part of a common class of phytochemical elements known as flavonoids, the anthocyanins are responsible for the red, purple, and blue colors found in many vegetables, flowers, and fruits. High consumption of fruits and vegetables is more and more related to the reduction of cancer and cardiovascular diseases, and anthocyanins can contribute to such effects. The beneficial effects of fruits with high contents of anthocyanins could be related to their antioxidant activities. However, it is still unknown if the consumption of anthocyanins contained in food significantly increases the serum antioxidant capacity in vivo.[1] Anthocyanins' bioavailability has been extremely low, and the proportion absorbed may be <0.1% of the ingested dose.[2] Many studies have used purified anthocyanin extract, and few have investigated the whole food effects in the absorption of anthocyanins and antioxidant activity. Anthocyanins are rarely ingested in isolated form, but they are currently consumed in combination with other food matrices, which generally are rich in fibers, and other flavonoids.[1] Besides the antioxidant activity of anthocyanins demonstrated in vitro, their possible effects in vivo mostly depend on absorption, metabolism, distribution throughout tissues, and excretion. However, there is considerable uncertainty about the precise mechanism of absorption of phenolics found in the diet by the gastrointestinal tract. However, it is known that anthocyanins can be present in blood, in both their glycosylated and aglycone forms. Nevertheless, from the nutritional point of view, the mere presence of a bioactive compound in food does not guarantee its biological effect. It is important to investigate the compound's stability in the gastrointestinal tract (absorption), distribution, metabolism, and biomarkers for possible biological actions. Thus, the antioxidant activity would be the first step to investigate some possible biological activities in vivo for the fruit evaluated in this study.[3]

Diverse techniques are currently used to measure the antioxidant activity of several compounds and foods; among them, the oxygen radical absorbance capacity (ORAC) assay should be emphasized. Ferric reducing ability (FRAP) and Trolox equivalence assay (TEAC) are other important techniques. These assays are based on different mechanisms that use different sources of free radicals or oxidants; consequently, these test results are not always directly correlated with one another.[4] The ORAC assay is the antioxidant preferable method; therefore, it is widely applied for the antioxidant capacity in human and animal plasma, proteins, and DNA, besides pure substances, plants, and food extracts.[5] ORAC methodology may be a good simulation of the physiologic conditions, such as blood temperature and pH. The ORAC assay uses peroxyl radicals, partially similar to some of the radicals in the biological systems, and could be refereed as the only antioxidant method that combines time and degree of inhibition in the evaluation of antioxidant capacity.[6] The TEAC technique has also been effective in determining the antioxidant activity of foods and even in the blood plasma of rats and humans because it is a simple method that, different from ORAC, is based on electron transfer reactions[7] and, therefore, was one of the methods selected for our study.

Jabuticaba is a berry from southeastern Brazil. *Myrciaria caulifolia* (Vell) Berg and *Myrciaria caulifolia* (DC) Berg are the varieties more suitable for "in natura" consumption as well as for food industry applications.[8,9] Berry diameter is about 3−4 cm; it has one to four seeds inside, and the peel is very purple. The pulp is white and sweet. Jabuticabas are very popular in

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Brazil and are most often consumed "in natura". Normally, the peel is not consumed. The berry can be used for jelly, jam, liqueur, and candy. The fruit is rich in anthocyanins: about 315 mg of total anthocyanins/100 g of fruit was determined using spectrophotometric methods.[10] Reynertson found 433 mg of cyanidin-3-glycoside and 81 mg of delphinidin-3-glycoside per 100 g of dry weight.[11]

Thus, the purpose of this study was verifying if the consumption of jaboticaba freeze-dried peel powder in different proportions in the diet could result in antioxidant effects in rat plasma.

MATERIALS AND METHODS

Fruit. Jaboticaba fruits (*Myrciaria jaboticaba* Vell Berg) were bought at a local market in Campinas in September 2008. The fruits were washed and manually peeled. The peels were frozen at −18 °C.[12,13]

Preparation of Freeze-Dried Jaboticaba Peels. The peels previously frozen at −18 °C were lyophilized in a Liobras freeze-dryer. The lyophilized product (jaboticaba peel powder) was stored at −80 °C.

Determination of Macronutrients. The content of protein was determined by Kjeldahl;[14] humidity and ashes were determined according to the method of the Adolfo Lutz Institute[15] and lipids according to the Bligh and Dyer method.[16] Total sugars content was determined using the Lane and Eynon[17] method.

Extraction of Anthocyanins for Mass Spectrometry and High-Performance Liquid Chromatography (HPLC) Analysis. One gram of freeze-dried jaboticaba peel was weighed and extracted with 15 mL of methanol/water/acetic acid (85:15:0.5, v/v, MeOH). The sample was then shaken in a vortex for 30 s and kept in an ultrasound for 5 min. The tube was then centrifuged at 4550*g* for 10 min, and the supernatant was removed. The sample was extracted once again with 10 mL of MeOH/H_2O/AcAc by using the same procedure. Supernatants were combined and filtered (0.22 μm Teflon filter).[18]

Analysis of Anthocyanins by Mass Spectrometry. Five microliters of the extract (see Determination of Macronutrients) was diluted in 995 μL of methanol/H_2O (1:1) and 0.1% of formic acid. For ESI, a Q-TOF mass spectrometer (Micromass, Manchester, U.K.) was used. The general conditions were as follows: source temperature of 100 °C, capillary voltage of 3 kV, and cone voltage of 35 V. The measurements were performed by direct infusion with a flow rate of 10 μL min^{-1}, using a syringe pump (Harvard Apparatus). Mass spectra were acquired and accumulated over 60 s, and spectra were scanned in the range between m/z 100 and 1000.

Analysis of Anthocyanins by High-Performance Liquid Chromatography Coupled to Photodiode Array (HPLC-PDA). The HPLC method for quantification of anthocyanin was based on that of Favaro[19] with slight modifications. The analyses were carried out in a HPLC (Waters 2996) system equipped with a photodiode array detector (Waters 515), software Empower. The separation of anthocyanins was done with a C18 column (ODS2, length = 25.0 cm, internal diameter = 4.6 mm, particle size = 5 μm, and pore size = 100 Å, Varian, Microsorb MV). The mobile phase used was deionized water/acetonitrile/formic acid (81:9:10 v/v/v). Isocratic elution with mobile phase flow volume of 1.0 m L min^{-1} was used; the injection volume was 20 μL; time of elution was 40 min, and detection was done at 520, 525, and 540 nm. Peak identification for each anthocyanin was based on the comparison of relative retention time (t_R), peak area percentage, and spectral data with anthocyanin standards. For anthocyanin quantification, cyanidin-3-*O*-glucoside, cyanidin-3-*O*-galactoside, and delphinidin 3-*O*-glycoside authentic standards (Extrasynthese) were used. The standard curves for all anthocyanins were determined by using the same chromatographic conditions described above.

Biological Assays: Administration of Jaboticaba Peel Powder to Rats. *Kinetics Tests.* The test was performed with 35 Wistar SPF (Specific Pathogen Free) adult rats, weighing approximately 250 g, from UNICAMP Biotherium Center (CEMIB), distributed in 5 groups of 7 animals each, with administration of commercial feed during the 7 first days for adaptation and later division of the groups. The animals were kept under controlled temperature (22 ± 2 °C) during the whole experiment, with alternate periods of 12 h in light and dark.

After a fast of 24 h,[19] the rats were weighed to calculate the amount of jaboticaba peel powder to be given to each one. The powder was dissolved in distilled water in such a concentration that provided ingestion of 7 mg of anthocyanins/100 g of corporal weight. The aqueous solution was administered by gavage. For determining the absorption kinetics, each group was killed within 0 15, 30, 60, and 120 min after the administration of freeze-dried jaboticaba peel powder solution, by means of decapitation, whereas the control group received water by gavage. The plasma was obtained by centrifugation of the blood and stored in a biofreezer (−80 °C) for later analyses.

The experiment was performed in conformity with the Ethical Principles for Experiments with Animals adopted by the Brazilian College for Animal Experimentation (COBEA), and it was approved by the Committee for Ethics in Animal Research − CEE/Unicamp − University of Campinas, Brazil (Protocol 1628-1).

Study of the In Vivo Antioxidant Activity of Freeze-Dried Powder of Jaboticaba Peel. In this test, 32 Wistar SPF adult rats (individual weight of approximately 250 g) were divided into 4 groups of 8 animals each. The animals were kept into individual cages and received AIN-93G,[21] added with jaboticaba peel powder. The five groups (N = 8) were divided according to the amount of jaboticaba peel added to the feed: 0 (control), 10, 20, and 40 g of jaboticaba peel powder/kg, as shown in Table 1 (the groups were denominated G0, the control group; G1, which received 1%, G2, which received 2%, and G4, which received 4% of freeze-dried powder of jaboticaba peel per kg in the diet). Sufficient amounts of sucrose were added to the diet, so as to adjust sugar levels at the same concentration found in the freeze-dried jaboticaba peel powder (Table 1).[22] The experiment took 28 days for all groups. The animals were kept under controlled temperature (22 ± 2 °C) during the whole experiment, with alternate periods of 12 h in light and dark. All animals were weighed every 2 days, and the diet consumed was monitored. Afterward, the animals were killed by decapitation, the blood was collected in heparin-coated tubes, and the plasma was obtained by centrifugation. The plasma samples were stored at −80 °C for later analyses of antioxidant activity. The experiment was performed in conformity with the Ethical Principles for Experiments with Animals adopted by the Brazilian College for Animal Experimentation (COBEA), and it was approved by the Committee for Ethics in

Table 1. Composition of the Experimental Diet[a]

ingredient	quantity, g/kg of food			
freeze-dried jaboticaba peel powder	0	10	20	40
starch	465.7	465.7	465.7	465.7
dextrin	155	155	155	155
casein	142	142	142	142
sugar	100	93.1	86.2	72.4
fiber	50	50	50	50
soy oil	40	40	40	40
mineral mix	35	35	35	35
vitamin mix	10	10	10	10
L-cystine	1.8	1.8	1.8	1.8
choline bitartarate	2.5	2.5	2.5	2.5
di-*tert*-butylhydroxytoluene (BHT)	0.008	0.008	0.008	0.008

[a] The table shows the composition of the four diets given to the rats during the experiments.

Table 2. Centesimal Composition of Freeze-Dried Jaboticaba Peel Powder

component	content (%)	SD
water	22.72	0.23
lipids	1.27	0.07
ashes	3.01	0.07
raw protein	3.90	0.12
raw fiber	6.45	0.28
reducing sugars	40.95	0.57
total reducing sugars	59.04	0.99
saccharose	18.09	ND

Animal Research − CEE/Unicamp − University of Campinas, Brazil (Protocol 1627-1).

Preparation of Plasma Samples for Antioxidant Tests. Plasma samples stored at −80 °C were slowly defrosted and shaken in a vortex. The methodology proposed by Prior et al.[23] was used, with some adaptations. Fifty microliters of plasma was transferred to an Eppendorf, and 100 μL of ethanol and 50 μL of distilled water were added. The solution was shaken during 30 s in a vortex, and then 200 μL of metaphosphoric acid (0.75 mol L^{-1}) was added. Once again, the mixture was shaken in a vortex for 30 s and centrifuged during 5 min at 1400 rpm at 10 °C. The supernatant was removed and stored at −70 °C. For the ORAC analysis, 80 μL of supernatant material was diluted with 420 μL of phosphate buffer (0.075 M, pH 7.4).

ORAC Method. Twenty microliters of plasma solution (with a final concentration of 0.027 μL of plasma per μL of solution), 120 μL of fluorescein in potassium phosphate buffer (final concentration of 0.378 μg/mL, pH 7.4), and 60 μL of 2,2′-azobis(2-methylpropionamidine) dihydrochloride (AAPH) (108 mg mL^{-1} in water) were added in the microplate. The AAPH solution was prepared just before the analysis. For the blank, the extract was substituted by potassium phosphate buffer. As a standard, Trolox was used in potassium phosphate buffer (0.1, 1.0, 10, 25, 50, 80, and 100 μM). After the addition of all reagents to the plate, the fluorescence was read at every 1 min, for 80 min. The following filters were used: emission 520 nm and excitation 485 nm. Determinations were made in triplicate.[24]

ABTS Test. The ABTS test was performed on the basis of the method described by Rufino et al.[25] and Le et al.[26] with modifications. ABTS solution was prepared [2,2′-azinobis(3-ethylbenzotiazoline-6-sulfonate)] by mixing 5 mL of ABTS 7.0 mM solution and 88 μL of potassium persulfate 2.45 mM solution, which was left to react for 12 h, at room temperature in the dark. Then, Milli-Q water was added to the solution until an absorbance value of 0.700 (±0.05) at 754 nm with a Novostar BMG Labtech microplate reader was reached. The determination of sample absorbance was accomplished at room temperature, 23 ±1 °C, after 6 min of reaction. Trolox was used as standard antioxidant. For sample preparation, the plasma solution (final concentration of 0.125 μL of plasma per μL of solution) was used. On a 96-well transparent microplate, 250 μL of ABTS solution and 50 μL of plasma solution were added, and the absorbance was read.

Statistics. ANOVA and regression analysis were carried out. A 5% significance level was used. Microsoft Excel,[27] SAS,[28] and Sisvar[29] were used.

RESULTS AND DISCUSSION

Fruit Characterization. The peel represented 28.9 ± 4.92% (m/m) and the pulp and seeds sum 66.82 ± 4.92% (m/m) of total fruit weight. The average gross weight of fruit was 5.63 g ± 1.00, and the lyophilization process average yield was 20.5% (freeze-dried mass/initial mass × 100%).

The results of the proximate composition of freeze-dried powder of jaboticaba peel are shown in Table 2. The data show that the powder is constituted mostly of sugars and water and, in smaller proportions, fibers and ash. Donadio[8] found similar values for the whole fruit, being 3.4% ashes, 4% raw protein, and 4.52% raw fiber. The other components, mostly water and sugars, showed different results as the author worked with the whole fresh fruit.

The chromatographic analysis of anthocyanins in the freeze-dried peel of the fruit showed the presence of delphinidin-3-glycoside and cyanidin-3- glycoside (Figure 1). This was confirmed by comparing retention times with the authentic standards and also by chromatography coupled to mass spectrometry (LC-ESI-MS/MS Q-ToF) (Table 3).

The total content of anthocyanins analyzed by HPLC was 2599.3 mg 100 g^{-1} of freeze-dried powder. Cyanidin 3-glycoside (peak 2) was the dominant anthocyanin (75.6% of total anthocyanins).

Rats' Weight Gain. Considering the animals that ingested experimental diet, there was no significant statistical difference between the ingestion of food at G0, G1, G2, and G4. Table 4 shows the ingestion of anthocyanins in milligrams per 100 g of weight. The analyses indicated there was no difference in the gain of weight among animals that did not consume the powder and those that consumed diets with different amounts of jaboticaba powder. Moreover, no statistical difference was observed in the liver weight of the group control and the experimental groups. The same result was also found by Tsuda,[30] who worked with rats fed diets with added purified cyanidin 3-*O*-β-D-glucoside (95%), the same anthocyanin predominant in jaboticaba.

Kinetics Tests. There was no difference among concentrations applied for ABTS and ORAC variable responses for any of the times applied, 0, 15, 30, 60, and 120 min, considering an ingestion of 7 mg/100 g of animal weight. The model parameters were not significant in either linear or quadratic effects. Suda et al.,[20] working with the absorption of acylated anthocyanins originated from purple-fleshed sweet potato, found a 1.5-fold increase in the plasmatic antioxidant activity of rats 30 min after ingestion by gavage of 14.63 mg of peonidin 3-caffeoylsophoroside-5 glucoside equivalent/100 g of body weight. In this case, it is suggested that the dose was not enough to cause any verifiable effects.

In Vivo Antioxidant Activity of Freeze-Dried Powder of Jaboticaba Peel. *ABTS Tests.* The results of ABTS analysis for the plasma of rats treated with diet with jaboticaba peels added at different concentrations are presented in Figure 2. According to Figure 2, one should realize an increasing quadratic tendency; in other words, as the freeze-dried jaboticaba peel powder concentration added to the diet increases, the ABTS levels increase until 2% of jaboticaba powder is added to diet. From that point the values start to decrease, even though the concentration levels are increasing. The ideal dose for that kind of study is 23 g of freeze-dried jaboticaba peel kg^{-1} of food, and the estimated TEAC value is 22.78 Trolox equiv μL^{-1} according to the model adjusted. In this case, the antioxidant capacity has increased 1.7 times when compared to the group control.

ORAC Test. The coefficient of variation obtained was considerably satisfactory, indicating good precision for the experiment (R^2 = 99.69%), as shown in Figure 3.

A quadratic tendency in the antioxidant response as a function of the concentration of freeze-dried jaboticaba peels in the diets (significance of 0.05%) was observed. The dose used to reach

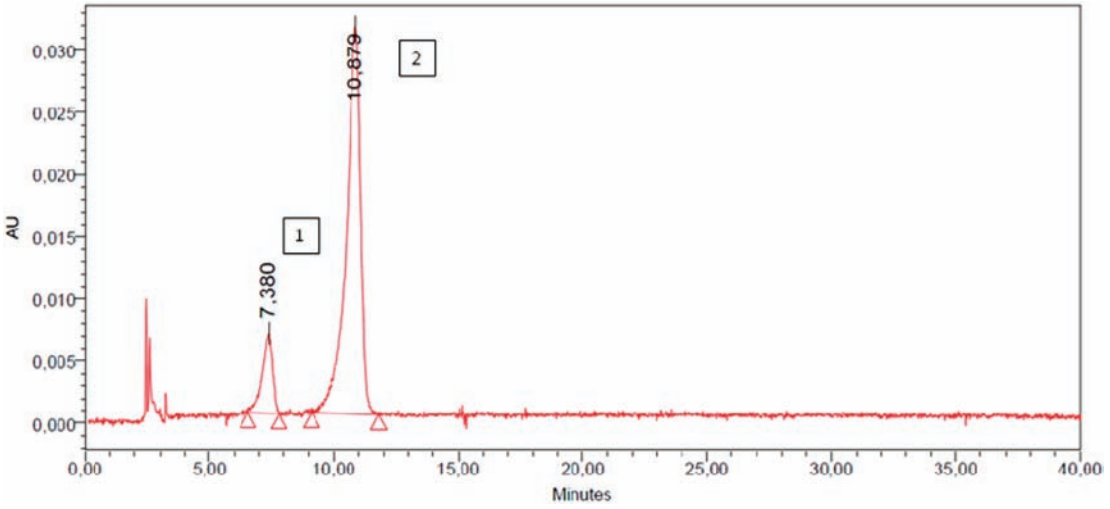

Figure 1. Chromatogram obtained from freeze-dried jaboticaba peel powder. Chromatographic conditions: column, C18; mobile phase, deionized water/acetonitrile/formic acid 81:9:10 (v/v/v); injection volume, 20 μL; and spectrophotometer detection at 520, 525, and 540 nm. Peak 1 refers to delphinidin-3-*O*-glycoside, 635.3 mg/100 g of jaboticaba peel (dry weight), whereas peak 2 is cyanidin-3-*O*-glycoside, 1964 mg/100 g of jaboticaba peel (dry weight).

Table 3. Identification of Anthocyanins from Jaboticaba Peels

					mg 100 g^{-1} of weight	
peak	t_R(min)	$[M]^+$ (m/z)	MS/MS (m/z)	anthocyanin	dry	fresh
1	7.38	465	303	delphinidin 3-*O*-glycoside	635.3	130.2
2	10.879	449	287	cyanidin 3-*O*-glycoside	1964	402.6
total					2599.3	532.9

Table 4. Ingestion of Diet and Anthocyanins[a]

	ingestion of diet		ingestion of jaboticaba peel		ingestion of anthocyanins	
group	g/day	DP	mg/day	DP	mg/100 g of body weight	DP
GO	21.2	1.4	0	0	0	0
G1	21.4	1.1	214	11	1.57	0.13
G2	22.8	1.5	456	30	3.36	0.13
G3	22.1	1.6	884	63	6.53	0.35

[a] Table shows the ingestion of diet for each of the groups described (see Material and Methods). The ingestion of diet was not significantly different among the groups. Ingestion of anthocyanin, as expected, varied according to the amount of jabuticaba peel for each group. Similar values of total anthocyanin ingestion are also described by other authors.

maximum antioxidant capacity was 2% (20 g of freeze-dried jaboticaba peel kg^{-1} of food), with estimated ORAC plasmatic value of 22.96 Trolox equiv μL^{-1}. In this case, the antioxidant capacity increased 1.3 times when compared to the control group. Tsuda et al.,[30] when working with diets of purified cyanidin 3-*O*-β-D-glucoside for rats and using lipid peroxidation methodology in the plasma for analysis of antioxidant potential, concluded that the group fed cyanidin had an increase in resistance to serum oxidation. This study has also analyzed plasma physiologic antioxidants and cholesterol fractions and concluded that the antioxidant action of purified cyanidin 3-*O*-β-D-glucoside seems to be due to a direct antioxidant effect.

Mazza et al.[31] verified the increased antioxidant capacity in humans after the consumption of an experimental meal rich in fats and supplemented with freeze-dried powder of blueberries measured by ORAC; however, such potential did not increase when measured by the TEAC method. Cao et al.[32] also found an increase of the plasmatic antioxidant capacity by ORAC test when evaluating humans who ingested 10 portions of fruits and vegetables on a daily basis. The 3-fold increase in the antioxidant capacity of human plasma was also emphasized by Mertens-Talcott et al.[33] in work accomplished with açaí berry pulp.

Another study reported increased human plasma antioxidant capacity (DPPH and ORAC) after consumption of blackberry juice with milk and water. However, according to the ORAC results, when the initial time was compared to 4 h after consumption of blackberry juice with water, there was an increase of 23% in the antioxidant capacity.[3]

Hassiomotto et al.,[2] when working with the metabolism and antioxidant activity of wild mulberries in rats, reported that only

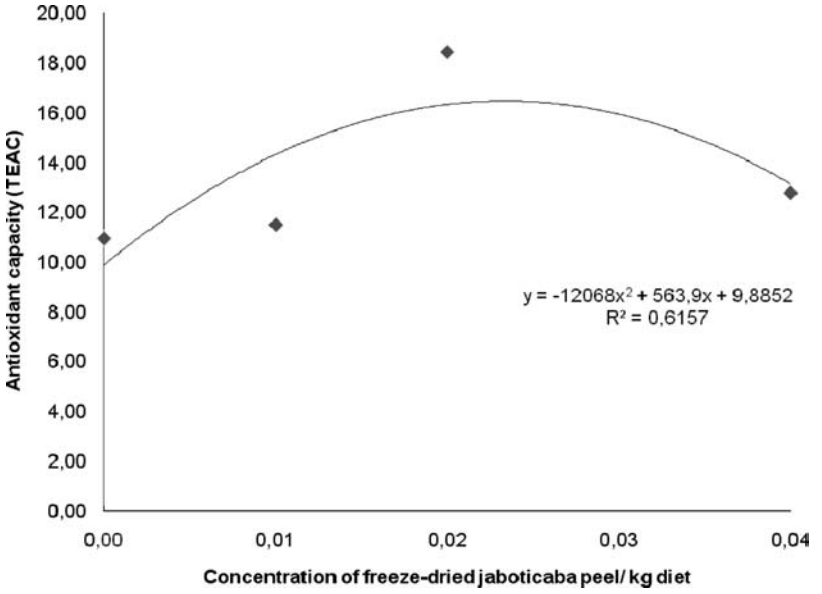

Figure 2. Relationship between concentrations of freeze-dried jaboticaba peel powder in the diet and antioxidant response by ABTS test. The figure shows the variation of plasmatic antioxidant capacity from the four groups investigated according to ABTS test. It is clear that group G2 (ingestion of diet with added 2% of jaboticaba peels) showed higher values for antioxidant capacity. Groups G1 and G4 showed antioxidant capacities similar to that of the control (G0).

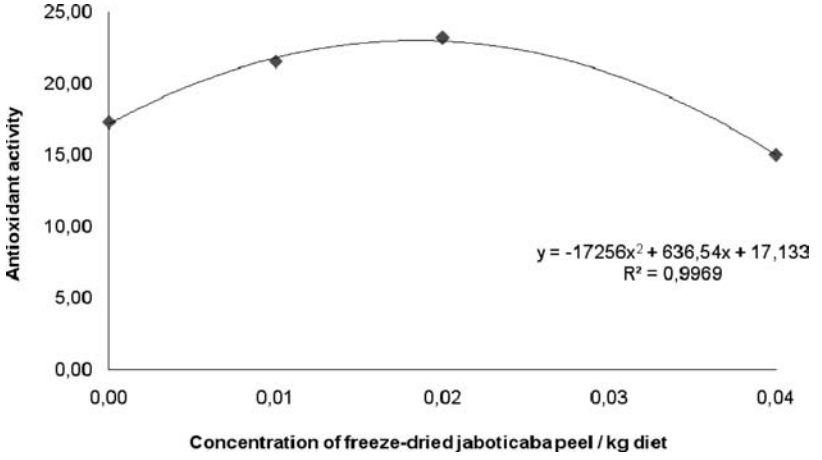

Figure 3. Relationship between concentrations of freeze-dried jaboticaba peel powder in the diet and antioxidant response by ORAC test. The figure shows the variation of plasmatic antioxidant activity from the four groups investigated according to ORAC test. Corroborating the results from the ABTS test, G2 showed the highest antioxidant response among the groups. Group G1 showed an antioxidant activity level between those of G0 (control) and G2. G4 showed smaller values of antioxidant capacity (even smaller than those of G0).

0.11% of the total anthocyanin ingested was absorbed; however, it was enough to increase the antioxidant capacity of the plasma evaluated by the linoleic acid oxidation method. The maximum value of antioxidant activity verified was reached after 15 and 30 min of the administration of extract rich in anthocyanin, with 44 ± 5 and $40 \pm 5\%$ of inhibition, respectively, corresponding to the highest value of total cyanidin concentration in the plasma. From 1 to 4 h after administration, the plasma antioxidant capacity decreased, but it was still higher ($\pm 30\%$) than the basal level Suda et al.,[20] who also evaluated the antioxidant activity in the plasma of rats that received concentrated extract of purple-fleshed sweet potato and found an increase in the plasmatic antioxidant potential (1.5 times more) at 30 min after administration. Such an increase was directly related with the anthocyanins identified in the plasma.

Walton et al.,[1] when working with concentrated powder of anthocyanin (32.9% of purity) in pigs, observed that the increased antioxidant activity due to anthocyanin ingestion was detected only for the FRAP test, but not for the ORAC test, different from the results presented in this study; perhaps it could be due to the different animal models used.

In our investigation, for both antioxidant tests used, ABTS and ORAC, the group that received a concentration of 4% of freeze-dried jaboticaba peel kg^{-1} of food did not present any antioxidant effect in the plasma compared to the control group. Thus, we conclude that the phenolic compounds can inhibit the oxidation processes in certain systems, but the dosage seems to have upper and lower limits. Some antioxidant compounds may present a pro-oxidizer activity under some conditions and concentrations.[34] Human cells are generally at reduced state,

but some degree of localized oxidation is necessary for important functions, so that the overload of reducer antioxidants may adversely affect their regular functions. A surplus of antioxidants may lead to the inhibition of cellular proliferation, once it prevents the transitory state of oxidation and decreases the adaptation to oxidative stress. It may also reduce free transition metals that turn into powerful catalysts of chain reactions started by free radicals.[35] Although it is still unclear, some authors speculate that excessive amounts of antioxidants could decrease the reactive oxygen species level, inhibit apoptosis, and suppress the elimination of cancer cells induced by anticancer drugs.[36]

Absorption of anthocyanins from jaboticaba is also affected by the matix of the food, so fiber is a major compound in plant foods, and many polyphenols can be associated with it on a food matrix. When this fiber−polyphenol complex is fermented by colonic microbiota, the release and absorption of some compounds or metabolites could occur, causing possible benefits for the consumer, especially in the antioxidant status.[37] Some points should be noted to consider the effectiveness of such compounds: concentration in the food and bioavailability in the food matrix. As considered above, the content of indigestible dietary fraction in food tends to be significantly greater than the fiber content; the indigestible fraction considers fiber, minerals, resistant starch, protein, and indigestible associated compounds other than dietary fiber.[38] This is very important information about the functionality of the food because these substances can be fermented by the colonic microbiota and they have physiological effects similar to those of the dietary fiber.[39,40]

This study evidenced the significant increase ($p = 0.05$) in the antioxidant potential of plasma in rats treated with jaboticaba peel. The results show that the excessive ingestion of anthocyanins from jaboticaba peels may lead to a decrease of the antioxidant activity, which may suggest that the recommended dietary intake of anthocyanins (or phenolic compounds) should be well established for healthy individuals.

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ABBREVIATIONS USED

ORAC, oxygen radical absorbance capacity; TEAC, Trolox equivalence assay; FRAP, ferric reducing ability; ABTS, 2,2′-azinobis(3-ethylbenzthiazoline-6-sulfonic acid); DPPH, 2,2-diphenyl-1-picrylhydrazyl radical; pH, potential hydrogen ionic; SD, standard deviation; ND, not determined.

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