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Artigo

Avaliação da Bioacessibilidade *in vitro* de Cobre, Ferro e Manganes em Frutas da Amazônia

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Evaluation of *in vitro* Bioaccessibility of Copper, Iron and Manganese in Amazonian Fruits

Abstract: The bioaccessibility of Cu, Fe and Mn in Amazonian fruits (açaí, white açaí, bacuri and tucumã) was evaluated in this study using *in vitro* methods. Flame atomic absorption spectrometry (FAAS) was used for total metal quantification, and graphite furnace atomic absorption spectrometry (GFAAS) was used to quantify the metals in the simulated gastric and gastrointestinal digestions. The Cu concentration in the fruits tested varied between 8.59 and 14.99 mg/kg. For Fe, the concentration in the samples ranged from 35.88 to 60.45 mg/kg. The lowest and highest concentrations of Mn were found in bacuri (5.01 mg/kg) and white açaí (709.93 mg/kg), respectively. In the açaí, white açaí and tucumã samples, the bioaccessible percentage of Cu was higher than the human can absorb. The available Fe value determined in the gastric simulation was similar to that observed in the gastrointestinal simulation, varying from 42.95 to 56.22%. The bioaccessible Mn fraction varied between 28.72 and 68.78% in the gastric simulation and between 26.81 and 64.85% in the gastrointestinal simulation. This study contributes to our knowledge of the actual amount of Cu, Fe and Mn absorbed by ingestion of the studied fruits.

Keywords: Bioaccessibility; Amazonian fruits; inorganic elements; FAAS, GFAAS.

Resumo

A bioacessibilidade de Cu, Fe e Mn em frutas da Amazônia (açaí, açaí branco, bacuri e tucumã) foram avaliadas neste estudo utilizando métodos *in vitro*. A espectrometria de absorção atômica com chama (FAAS) foi utilizada para a quantificação total de metal e a espectrometria de absorção atômica em forno de grafite (GF AAS) foi utilizada para quantificar os metais nas digestões gástricas e gastrointestinais simuladas. A concentração de Cu nos frutos testados variou entre 8,59-14,99 mg/kg. Para Fe, a concentração nas amostras variou de 35,88-60,45 mg/kg. As menores e maiores concentrações de Mn foram encontradas nos bacuri (5,01 mg/kg) e açaí branco (709,93 mg/kg), respectivamente. Nas amostras de açaí, açaí branco e tucumã, o percentual bioacessível de Cu foi maior do que o ser humano pode absorver. O valor de Fe disponível na simulação gástrica foi semelhante ao observado na simulação gastrointestinal, variando de 42,95 a 56,22%. A fração Mn bioacessível variou entre 28,72-68,78% na simulação gástrica e 26,81-64,85% na simulação gastrointestinal. Este estudo contribui para o conhecimento da quantidade real de Cu, Fe e Mn absorvidos pela ingestão dos frutos estudados.

Palavras-chave: Bioacessibilidade; Frutas da Amazônia; Elementos inorgânicos; FAAS; GFAAS.

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Avaliação da Bioacessibilidade *in vitro* de Cobre, Ferro e Manganes em Frutas da Amazônia

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1. Introduction

The agriculture of the Amazonian region is based on cultivation of vegetables, native roots, herbs and exotic fruits. The fruits from this region are widely consumed, but are better known for their flavor than for their nutritional properties. Some native or non-native fruits species are a little consumed, but have a high nutritional potential, such as açaí (*Euterpe olearacea*), white açaí (*Euterpe* sp.), bacuri (*Attalea phalerata*) and tucumã (*Astrocarium aculeatum*).[1-4]

Açaí palm has multiple uses, as the stipe (stem), roots, leaves, palm and fruits can all be utilized. Açaí has several morphological characteristics; for example the white açaí, which is evident from its dark green color.[5,6] "Açaí wine" is extracted from the fruit, and is the main product produced from açaí. It has essential nutritional properties and is important for human heath, in addition to being a source of energy, fiber, anthocyanins and minerals. It has also been associated with the prevention of degenerative diseases.[7]

Bacuri has a white soft pulp with a sweet flavor, and is also considered to have a strong exotic aroma. It contains several types of proteins, vitamins and minerals, a high concentration of carotenoids and also possesses antioxidant activity.[1,8,9]

Tucumã is species of palm with edible pulp and seeds. The fruit is a source of carotenoids and vitamin A. The level of vitamin A found in tucumã is superior to other fruits, including papaya and acerola. Bioactive compounds are also found in this fruit.[1,10,11]

Several inorganic elements are considered to be essential for humans, however, their absorption is rather complicated. In this way the determination of total food content ingested only is not sufficient to create an absorption efficiency profile.[12]

Copper absorption in humans ranges from 25 to 70%, and the efficiency of absorption is directly linked to the nutritional status of the individual. Copper is an essential micronutrient for living organisms, as it is a constituent of blood, it participates in several processes in the body, and it is a component of many proteins and enzymes.[12-14]

The absorption of iron occurs primarily in the bowel and, according to Grotto (2008), approximately 1 to 2 mg of iron is absorbed per day via the duodenum. Iron is considered to be essential, as it is required for several critical functions in the human body. Almost all iron in the body is found within cells or bound to hemoglobin or myoglobin.[12,16,17]

Only a small proportion of ingested manganese is absorbed by humans, independent of the amount consumed. Manganese is present in tissues rich in mitochondria, where it is required for the formation of connective tissue, skeletal muscle, cartilage and bone, in addition to its antioxidant role in the body.[12,18]

Bioaccessibility studies evaluate the fraction of a compound released from the matrix of a particular food in the gastrointestinal tract that can be absorbed by an organism. Bioaccessibility is an indicator of the bioavailability of food, which can be used to determine the maximum amount of a compound that may reach the systemic circulation.[12,19,20] Kulkarni, Acharya, Rajurkar and Reddy (2007) evaluated the bioaccessibility of essential elements in a wheatgrass sample, and reported bioaccessibility levels ranging from 9 to 60%. Nascimento, Naozuka and Oliveira (2010) analyzed the bioaccessible fraction of Cu and Fe in a cashew nut sample, and reported values of 83 and 78%, respectively. Lima, Soares, Silva, Figueiredo, Sousa and Menezes (2014) studied the *in vitro* bioaccessibility of Cu, Fe and Zn in cashew apple juice and cashew apple fiber, and the results showed that the bioaccessible fractions in the juice varied from 4–15%, which was less than 5% in the fiber. These studies demonstrate the importance of evaluating the bioaccessibility of inorganic elements, as the bioaccessible fraction depends on the matrix used in the study.

In the present study we used spectrometric techniques to determine Cu, Fe and Mn in digested samples and bioaccessible fractions of fruits after simulated *in vitro* gastric and gastrointestinal digestion.

2. Materials and methods

2.1. Instrumentation

The fruit samples were dried using a lyophilizer (L101, Liotop, São Carlos, Brazil). The digestion of the samples was performed using a microwave oven (START E, Milestone, Sorisole, Italy).

The determination of Cu, Fe and Mn in the digested samples was performed using a flame atomic absorption spectrometer (FAAS, iCE 3300, Thermo Scientific, Cambridge, United Kingdom). Hollow cathode lamps (Photron Pty. Ltd., Victoria, Australia) were used as the radiation source for Cu, Fe and Mn, operating at 4.0, 6.0 and 5.0 mA,

respectively. The wavelengths used were 324.8, 248.3 and 279.5 nm and the spectral resolution was 0.5, 0.2 and 0.2 nm for Cu, Fe and Mn, respectively. An air-acetylene mixture was used as the oxidant and fuel gas, respectively.

The levels of Cu, Fe and Mn in the bioaccessible fractions were determined using a graphite furnace atomic absorption spectrometer (SpectrAA 240Z, Varian, Victoria, Australia) equipped with an automatic sampler and background broker with Zeeman effect. Hollow cathode lamps were used as the radiation source. Argon of 99.999% purity (Linde, Pará, Brazil) was used as the gas purge for all steps of the heating program of the graphite furnace, except in the atomization step. The heating program and instrument parameters used for GFAAS are shown in Table 1.

Table 1. Instrumental parameters for determination of Cu, Fe and Mn in bioaccessible fractions by GFAAS.

Element	Wavelength (nm)			Lamp current (mA)			Spectral resolution (nm)		
Cu	327.4			0.5			0.5		
Fe	386.0			11.0			0.2		
Mn	279.5			4.0			0.2		
Step	Temperature (°C)			Time (s) (Ramp, Hold)			Gas flow (L/min)		
	Cu	Fe	Mn	Cu	Fe	Mn	Cu	Fe	Mn
1	95	90	95	5; 10	5; 15	5; 10	3.0	3.0	3.0
2	120	120	120	15; 15	10; 30	20; 15	3.0	3.0	3.0
3	900	1200	800	5; 5	5; 5	5; 3	3.0	3.0	3.0
4	2300	2300	2400	0.7; 2	0.5; 2	0.8; 2	0	0	0
5	2400	2400	2500	1; 2	1; 3	1; 2	3.0	3.0	3.0

A thermostatic bath (Q226M2, Dubnoff, Quimis, São Paulo, Brazil) was used for the *in vitro* gastric and gastrointestinal digestion simulations. A centrifuge (2K15, Sigma, Munich, Germany) was used to separate the phases after *in vitro* gastrointestinal digestion.

2.2. Reagents

All reagents used were of analytical grade. The solutions were prepared with distilled-deionized water (resistivity of 18.2 MΩ cm) from an ELGA water purification system (Elgastat, UK).

For the acid digestion we used 65% v/v HNO_3 (Sigma-Aldrich, St. Louis, MO, USA) and 30% w/w H_2O_2 (Quimex, São Paulo, Brazil). The gastrointestinal digestion was prepared using pepsin, pancreatin, α-amylase and bile salts (Sigma-Aldrich), NaOH (Synth, São Paulo, Brazil), HCl (Quimex) and NaCl (Synth).

Stock standard solutions of 1000 mg/L of Cu, Fe and Mn (Specsol, São Paulo, Brazil) were used to prepare the reference analytical solutions.

2.3. Samples

The açaí and white açaí pulps were purchased at a local street market in Belém (Pará State, Brazil). The bacuri and tucumã samples were obtained from the Ver-o-Peso market hall (traditional vendor of local fruits, vegetables and fish in Belém). The bark and seeds of the bacuri and tucumã were removed, and only the fruit pulp was studied. The pulp of each fruit was packaged and frozen at –20 ºC, and the samples were lyophilized.

2.4. Elemental analysis

Approximately 0.25 g of each sample was weighed (n = 3) and digested with 4 mL of 7.0 mol/L HNO_3 and 4 mL of 30% w/w H_2O_2. The heating program was performed in two steps, the first step was performed for 10 min at 200 ºC and 800 W, and the second step was performed for 15 min at 200 ºC and 800 W. The third step cooled the system through forced ventilation for a period of 50 min. After digestion, the samples and blank solutions were transferred to volumetric flasks and the volume was adjusted to 10 mL with ultrapure water. The element concentrations were determined in digested by FAAS.

The analytical curves used for Cu, Fe and Mn quantification by FAAS were 1.0, 2.0, 3.0 and 4.0 mg/L for Cu, 2.0, 4.0, 6.0 and 8.0 mg/L for Fe and 1.0, 2.0, 3.0 and 4.0 mg/L for Mn. The limits of detection (LOD) and limits of quantification (LOQ) were calculated from the equations $3 \times s/b$ and $10 \times s/b$, where s is the standard deviation of 10 measurements of the analytical blank and b is the slope of the analytical curve.

2.5. Simulated *in vitro* gastric and gastrointestinal digestion

The bioaccessibility of Cu, Fe and Mn in the Amazonian fruits was evaluated using the *in vitro* procedure described by Khouzam, Pohl and Lobinski (2011) and Moreda-Piñeiro et al. (2012), with adaptations. The *in vitro* gastric and gastrointestinal digestion of fruits were performed using gastric fluid (GF) and intestinal fluid (IF) solutions. The GF was prepared from 1% w/v pepsin in 0.15 mol/L NaCl, acidified with HCl to pH 2.5. The IF consisted of 3% m/v pancreatin, 1% m/v α-amylase and 1.5% m/v bile salts in 1.0 mol/L NaOH.

For the gastric digestion procedure, 0.3 g (n = 3) of each sample was placed in a 50 mL volumetric flask, to which 5 mL of GF was added. The mixture was agitated for 1 min for initial degassing, then heated in a thermostatic bath for 4 h at 37 ºC with constant agitation. The enzymatic reaction was stopped by placing the volumetric flasks in an ice bath for 10 min. After this, the solutions were centrifuged at 4000 rpm for 15 min. The supernatants were filtered using cellulose filters (0.45 µ membrane; Millipore, Bedford, MA, USA), then stored at 4 ºC. The supernatants obtained were acidified with HNO_3 for an acid final of 0.2 vol % for analysis. The concentrations of elements in the bioaccessible fractions were determined using GFAAS.

The gastrointestinal digestion procedure was performed after the gastric digestion, after the pH of the solution had been adjusted to 7.4 by the addition of an adequate volume of a 1.0 mol/L NaOH solution. A 5 mL aliquot of IF was added, and the resulting solution was stirred for 1 min, then incubated in a thermostatic bath at 37 ºC for 4 h with constant agitation. After incubation, the volumetric flasks were placed in an ice bath for 10 min, and the solutions centrifuged at 4000 rpm for 15 min. The supernatants were filtered using 0.45 µ membrane cellulose filters (Millipore, Bedford, MA, USA) and stored at 4 ºC. The supernatants obtained were acidified with HNO_3 for a final acid concentration of 0.2 % (v/v) for analysis. The concentrations of each of the elements in the bioaccessible fractions were determined by GFAAS.

The analytical curves used for Cu, Fe and Mn quantification by GFAAS were 6.0, 12.0, 18.0 and 24.0 µg/L for Cu; 3.0, 6.0, 9.0 and 12.0 µg/L for Fe; and 1.0, 2.0, 3.0, 4.0 and 5.0 µg/L for Mn. The limits of detection (LOD) and limits of quantification (LOQ) were calculated from the equations $3 \times s/b$ and $10 \times s/b$, where s is the standard deviation of 10 measurements of the analytical blank and b is the slope of the analytical curve.

The bioaccessibility of Cu, Fe and Mn is defined as the proportion of these elements available for absorption, [29] calculated as follows:

$$\mathrm{BF}\ (\%) = \frac{[\mathrm{M}]\mathrm{GD}}{[\mathrm{M}]\mathrm{AD}} x\ 100$$

where BF (%) is the percentage of bioaccessible metal, [M] is the metal concentration after the *in vitro* digestion procedure, and [M] is the metal concentration determined in the fruits after the acidic digestion process.

2.6. Method validation

The accuracy of the total element concentration measurements by FAAS and the simulated *in vitro* gastric and gastrointestinal digestion by GFAAS were evaluated using spike experiments. The digested samples were spiked with 0.5, 1.0 and 1.5 mg/L Cu; 2.0, 4.0 and 6.0 mg/L Fe; and 1.0, 2.0 and 2.5 mg/L Mn. The bioaccessible fractions were spiked with 8.0, 12.0 and 18.0 µg/L Cu; 2.0, 4.0 and 5.0 µg/L Fe; and 1.0, 1.5 and 2.5 µg/L Mn. The recovery (%) was defined by *observed value/expected value* × 100.[31]

3. Results and discussion

3.1. Total concentration of Cu, Fe and Mn in fruits by FAAS

The concentrations of Cu, Fe and Mn (mg/kg) in açaí, white açaí, bacuri and tucumã are presented in Table 2.

Table 2. Concentrations of Cu, Fe and Mn (mg/kg) in fruits by FAAS (n = 3)

Sample	Cu	Fe	Mn
Açaí	7.71 ± 0.33	60.45 ± 2.06	534.28 ± 2.01
White açaí	14.99 ± 0.77	59.64 ± 2.61	709.93 ± 9.35
Bacuri	13.79 ± 0.54	53.72 ± 6.73	5.01 ± 0.28
Tucumã	8.59 ± 0.48	35.88 ± 1.35	62.98 ± 1.83

The Cu concentration in the fruit samples varied in the range 8.59–14.99 mg/kg. The lowest Cu concentration was found in tucumã, and the highest concentration in white açaí. Other studies in the literature have reported Cu levels in different types of fruits, such as banana (0.946 mg/kg), jackfruit (11.78 mg/kg) and mango (7.891 mg/kg).[26] Previous studies have reported a concentration of 21.5 mg/kg Cu in açaí[5] and 6.8 mg/kg in bacuri-azedo.[27] For Fe, the concentration determined in the fruits evaluated in this study varied from 35.88–60.45 mg/kg. The Fe concentration was highest in açaí, followed by white açaí, bacuri, and finally, tucumã, which had the lowest Fe concentration. Menezes et al. (2008) reported Fe values in lyophilised açai pulp of 45 mg/kg, close to those reported in this study. Berto et al. (2015) obtained lower

levels of Fe (9.1–33 mg/kg) in various native Amazonian fruits (bacuri-azedo, biribá, cubiu, ingá-açu, pajurá, piquiá, sapota, umari and uxi) compared to the Fe content of the fruits evaluated in this study.

The lowest and highest Mn concentrations in the sampled fruits were found in bacuri (5.01 mg/kg) and white açaí (709.93 mg/kg), respectively. The higher levels of Mn obtained in this species of açaí may be related to their habitat, as white açaí grow in the "varzea region" (flood forest), and this can affect the balance of this element in environment.[28] When compared to previous studies that reported the Mn concentration in apples (0.82 mg/kg), fruits of the Amazonian region appear to contain higher levels of this metal.[24]

3.2. In vitro bioaccessibility of Cu, Fe and Mn in fruits by GFAAS

The concentrations and the bioaccessible fractions (%), obtained by the simulated *in vitro* gastric digestion of Cu, Fe and Mn in Amazonian fruits, are presented in Table 3.

Table 3. Concentrations (mg/kg) and the bioaccessible fractions (BF%) of Cu, Fe and Mn in fruits obtained by *in vitro* simulated gastric digestion (n = 3)

	Cu		Fe		Mn	
	Conc. (mg/kg)	BF%	Conc. (mg/kg)	BF%	Conc. (mg/kg)	BF%
Açaí	6.52 ± 0.09	85	27.25 ± 0.91	45	239.78 ± 34.65	45
White açaí	10.50 ± 0.41	71	33.53 ± 2.23	56	488.29 ± 28.08	69
Bacuri	6.28 ± 0.09	45	24.21 ± 1.48	45	1.44 ± 0.09	29
Tucumã	7.61 ± 0.36	88	16.23 ± 2.27	45	36.55 ± 0.39	58

The gastric digestion process was used to analyze the gastric fluid absorption in isolation. The bioaccessible concentration (mg/kg) and bioaccessible fraction (%), obtained by the *in vitro* simulated gastrointestinal digestion of Cu, Fe and Mn in Amazonian fruits (Table 4), were used to determine the accessible metal concentration and absorption of each metal by the human.

Table 4. Concentrations (mg/kg) and bioaccessible fractions (BF%) of Cu, Fe and Mn in fruits obtained by *in vitro* simulated gastrointestinal digestion (n = 3)

	Cu		Fe		Mn	
	Conc. (mg/kg)	FB %	Conc. (mg/kg)	FB %	Conc. (mg/kg)	FB %
Açaí	4.03 ± 0.26	52	25.96 ± 0.83	43	346.48 ± 20.27	65
White açaí	7.79 ± 0.15	53	32.55 ± 2.83	55	339.77 ± 2.27	48
Bacuri	3.79 ± 0.09	27	22.77 ± 1.68	42	1.84 ± 0.66	37
Tucumã	4.88 ± 0.23	57	19.72 ± 0.81	55	16.88 ± 0.08	27

According to Mahan and Escott-Stump (2005) and Fairweather-Tait (1992), Cu absorption varies between 25 and 70%, and is more efficiently absorbed in the intestine. As shown in Tables 4 and 5, the gastric digestion obtained a higher level of Cu

available for absorption. The bioaccessible Cu percentage in the açaí, white açaí and tucumã samples was higher than the amount of Cu that the organism can absorb. Therefore, this level of Cu will not be completely absorbed by the organism.

Some authors,[12,30] have suggested that the site of greatest Cu absorption in the gastrointestinal system is the duodenum. In the present study, the bioaccessible fraction determined in the gastrointestinal simulation was lower than the gastric simulation, which may be explained by the characteristics of the fruits, as fiber and phytate can slightly inhibit Cu absorption. As Cu is an essential nutrient widely distributed in foods, studies evaluating Cu bioaccessibility are relevant. Previous studies have reported a bioaccessible potential in foods including juices (15%), nuts (75%) and meat (40%) similar to that found in the fruits analyzed in this study, which varied from 27–88%. This bioaccessible potential fraction contributes the amount of Cu that the human body needs in order to perform various functions.[19,22,23]

In humans, the amount of iron absorbed is 25% in the form of heme iron and 10% for non-heme iron. According to Fairweather-Tait (1992) and Mahan and Escott-Stump (2005), absorption of combined heme and non-heme iron varies between 5 and 15%. Tables 4 and 5 show that the gastric and gastrointestinal simulations gave similar results regarding the bioaccessible levels in the fruits, which ranged from 42 to 56%.

The absorbed Fe content determined for the studied fruits is higher than the amount of Fe that can be absorbed by the organism. According to Mahan and Escott-Stump (2005), non-heme iron (Fe^{3+}) is the predominant form in samples of plant origin. Therefore, the absorption of Fe is less efficient, as iron is better absorbed in its reduced form (Fe^{2+}), as it is not necessary to form chelates with sugars, amino acids and vitamin C in order to be absorbed. Most of the samples (açaí, white açaí and bacuri) showed a higher percentage absorption in the gastric simulation. This can be explained by the acidity of the solution used for this simulation, as ferric iron precipitation occurs at basic pH. The gastrointestinal simulation was performed at pH 7; therefore, any iron that was not chelated became unavailable for absorption.[12] According to Mahan and Escott-Stump (2005), dry beans and vegetables are the best source of Fe in foods of plant origin. Previous studies have determined the availability of Fe in several other plant foods. Kulkarni et al. (2007) reported 31–34% of bioaccessible Fe in a wheatgrass sample. Nascimento et al. (2010) obtained a higher percentage of Fe (70%) in the bioaccessible fraction of cashew nuts compared the Fe content found in the fruits evaluated in the current study. However, Lima et al. (2014) reported a lower level of bioaccessible Fe (11%) in cashew apple juice.

Fairweather-Tait (1992) reported that the daily Mn absorption by the organism is between 3 and 4%. The bioaccessible fraction of Mn varied from 29–69% in the gastric simulation, and from 27–65% in the gastrointestinal simulation. The highest Mn concentration available for absorption was found in açaí (239.8 mg/kg in the gastric simulation and 346.5 mg/kg in the gastrointestinal simulation) and white açaí (488.3 mg/kg in the gastric simulation and 339.8 mg/kg in the gastrointestinal simulation). All fruits tested contained Mn at a concentration higher than the required daily intake. However, Mn concentration varies in foods, and foods of plant origin are considered to be good sources of this mineral. Khouzam et al. (2011) reported a higher Mn value in the bioaccessible fraction of apples (92%) compared to the bioaccessible Mn content obtained in the fruits included in the current study. However, we observed a higher bioaccessible percentage of Mn when compared to the values reported by Moreda-Piñeiro et al. (2012) in marine foodstuffs (5–22%).

3.1. Figures of merit

Table 5 shows the parameters used for the determination Cu, Fe and Mn by GFAAS and FAAS.

3.4. Accuracy

The accuracy was also evaluated by spike recovery tests by FAAS and GFAAS (Table 6). The samples were spiked with three different concentration levels of each analyte.

Table 5. Figures of merit obtained for determination of Cu, Fe and Mn by GFAAS and FAAS

	Cu	Fe	Mn
	GFAAS		
LOD (µg/L)	1.52	0.10	0.28
LOQ (µg/L)	5.07	0.34	0.95
SLOPE	0.0041	0.0054	0.0193
R^2	0.996	0.997	0.997
	FAAS		
LOD (mg/L)	0.01	0.17	0.02
LOQ (mg/L)	0.05	0.55	0.08
SLOPE	0.1107	0.0341	0.1373
R^2	0.999	0.996	0.991

LOD = limits of detection; LOQ = limits of quantification; R^2 = correlation coefficient

Table 6. Recoveries for the determination of Cu, Fe and Mn using spike experiments by FAAS and GFAAS

GFAAS	Spiked (µg L^{-1})	Found (µg L^{-1})	Recovery (%)
Cu	8.0	15.58	104
	12.0	15.64	118
	18.0	18.56	95
Fe	2.0	2.73	88
	4.0	4.40	95
	5.0	4.50	82
Mn	1.0	1.53	95
	1.5	3.93	95
	2.5	3.21	101
FAAS	Spiked (mg L^{-1})	Found (mg L^{-1})	Recovery (%)
Cu	0.5	0.55	110
	1.0	1.03	103
	1.5	1.6	107
Fe	2.0	2.2	110
	4.0	4.4	110
	6.0	6.7	112
Mn	1.0	1.01	101
	2.0	2.2	110
	2.5	2.48	99

The recoveries obtained by FAAS and GFAAS varied from 99 to 112% and 82 to 118%, RESPECTIVELY. These values are acceptable, and therefore, the measurement accuracy by FAAS and GFAAS is adequate.

4. Conclusions

Spectrometric techniques were used to determine the total concentration and bioaccessibility of Cu, Fe and Mn in Amazonian fruits. Açaí and white açaí contained the highest concentration of all inorganic elements tested, particularly Mn. For all samples, the highest absorption percentage in the bioaccessible fraction was obtained for Cu by the *in vitro* gastric digestion. The levels of Mn varied for the bioaccessible fractions obtained by gastric and gastrointestinal digestions. The percentage of Fe absorption was similar in all samples for the gastrointestinal digestion. The study of bioaccessibility allows for the determination of the actual amount of Cu, Fe and Mn absorbed in the body from consuming the studied fruits.

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