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CHEMICAL AND NUTRITIONAL EVALUATION OF KERNELS OF BOCAIUVA, *Acrocomia aculeata* (JACQ.) LODD.¹

Priscila A. HIANE², Paulo Aparecido BALDASSO³, Sérgio MARANGONI³, Maria Lígia R. MACEDO^{4,*}

SUMMARY

Protein characterization and results of proximate composition and mineral analyses of fruit kernels of bocaiuva, *Acrocomia aculeata* (Jacq.) Lodd., are reported. The kernels presented high contents of oil (51.7%), protein (17.6%) and fiber (15.8%). The seeds' soluble proteins were isolated according to their solubility. The main separated proteins were globulins (53.5%) and glutelins (40.0%). Moreover, the presence of low molecular mass proteases in these two fractions was shown by the SDS-PAGE method. The assays of protease-inhibitory and hemagglutinating activities showed that bocaiuva's protein fractions were not resistant to trypsin or chymotrypsin activities and that both had low lectin content. The globulin *in vitro* digestibility assay resembled a casein standard. Neither globulin nor glutelin enzymatic hydrolyses increased significantly ($p < 0.05$) after heat treatment. Threonine and lysine are the most limiting amino acids, respectively from two major protein fractions of the bocaiuva kernel, globulin (47.1% amino acid score) and glutelin (49.5% amino acid score), in terms of the theoretical profiles for children in the age range of 2 to 5 years recommended by the FAO/WHO. Bocaiuva kernels are found to be rich in calcium, phosphorus and manganese compared to some fruit nuts such as cashew and coconut.

Keywords: kernel, *Acrocomia aculeata*, globulin, glutelin, chemical analysis, digestibility.

RESUMO

AValiação QUÍMICA E NUTRICIONAL DE AMÊNDOAS DA BOCAIUVA, *Acrocomia aculeata* (JACQ.) LODD. Caracterização de proteínas e análises da composição centesimal e teores de minerais foram realizados nas amêndoas da bocaiuva, *Acrocomia aculeata* (Jacq.) Lodd. As amêndoas apresentaram alto teor de lipídio (51,7%), proteína (17,6%) e fibra (15,8%). Proteínas solúveis das sementes foram fracionadas de acordo com a sua solubilidade. As principais proteínas separadas foram as globulinas (53,5%) e glutelinas (40,0%), e a presença de proteases de baixo peso molecular nessas duas frações foi revelada por eletroforese em gel de poliacrilamida (SDS-PAGE). Ensaios da atividade inibitória de proteases e da hemaglutinação mostraram que as frações protéicas da bocaiuva não foram resistentes à ação da tripsina e quimotripsina e apresentaram baixo teor de lectina. A digestibilidade *in vitro* da globulina foi semelhante à da caseína padrão. Hidrólises enzimáticas da globulina e glutelina não aumentaram significativamente ($p < 0,05$), com o aquecimento. Treonina e lisina são os aminoácidos mais limitantes, respectivamente, das duas principais frações de proteínas da amêndoa da bocaiuva, a globulina (escore de aminoácido de 47,1%) e glutelina (escore de aminoácido de 49,5%), relativamente ao padrão teórico para crianças de 2 a 5 anos de idade, recomendado pela FAO/WHO. Amêndoas de bocaiuva mostraram ser ricas em cálcio, fósforo e manganês, em comparação com algumas amêndoas de frutos como caju e coco.

Palavras-chave: amêndoa, *Acrocomia aculeata*, globulina, glutelina, análise química, digestibilidade.

1 - INTRODUCTION

In the southwest of Brazil, Bocaiuva is the popular name for the palmtree *Acrocomia aculeata* (Jacq.) Lodd., Palmae, and its fruit. The species is native to the state of Mato Grosso do Sul, where it is particularly abundant. Other species of the genus occur not only in this state, but also in the southeast of Brazil as well as in Central America. In Brazil, the *Acrocomia* species are known by a number of common names, which include 'macauba', 'coco catarro', 'macabira', 'mocajuba' and 'macaiba'. Kernels of the bo-

caiúva fruit can be eaten raw or as candies such as 'paçoca' (replacing peanuts, the traditional main ingredient) and 'cocada' (replacing coconut) [2].

The cerrado region in Brazil is home to numerous fruit species, some of them with a potential in the market of fruit pulp and nuts, such as the *piqui*, *baru* and *jatobá-do-cerrado* species. Studies have been conducted with the purpose of supplying information that can promote commercial sales of such species and their larger use by the local population as supplementary sources of essential nutrients [14, 21, 27].

Bocaiuva kernels have a high protein content [3, 14], but data concerning their amino acid profile, antinutritional and/or toxic properties are not available.

Food proteins can be classified according to qualitative criteria based on their amino acid profiles and proportions and on their bioavailability and susceptibility to hydrolysis during digestion. Protein nutritional value is related to the quality of its amino acids [11]. Among the sources of plant protein, grains provide protein of high biological value, though deficient in some of the essential amino acids [30].

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The total protein fraction of seeds is a complex mixture of globulins (40-60%), albumins (8-20%), prolamins and glutelins, of which the first two constitute the main components. The proportions, however, differ among species, varieties and/or cultivars [7]. The literature reports that seed protein characterization and evaluation are central aspects in both basic and applied studies in nutrition and biotechnology areas [20, 33].

Storage proteins, such as lectins, ureases and proteolytic enzyme inhibitors correspond to 5% or more of the total protein present in seeds, reserve organs and other plant parts [7].

Some seed proteins such as lectins, proteinase inhibitors and vicilins have been extensively studied in Leguminosae (beans, soybeans, peanuts) and cereals (wheat, rye, barley). In plants, these proteins are associated to the defense mechanism and have bactericide, fungicide and insecticide properties [10, 19]. In human and animal diets, they are considered to play an antinutritional and/or toxic role, as they can inhibit growth in experimental animals, as well as decrease protein digestibility and cause pancreatic hypertrophy and hyperplasia [18]. The evaluation of protein fractions in foodstuffs can thus have a direct influence on the acceptability and use of food items as it can reveal characteristics that are related to their quality and safety in the context of Public Health and Nutrition [30].

In the present investigation, protein fractions of bocaiuva kernels were evaluated focusing on their nutritional potential by carrying out chemical analyses for proximal composition, amino acid composition, mineral content, activity as enzyme inhibitors and protein isolation and digestibility.

2 - MATERIAL AND METHODS

2.1 - Material

Mature fruits of the bocaiuva, *A. aculeata* (Jacq.) Lodd., were collected from the *campus* at the Universidade Federal de Mato Grosso do Sul (UFMS) in Campo Grande, MS, Brazil from December, 2002 to March, 2004 — except for February to April, which are months that are not part of the harvesting season.

The kernels were removed from the seeds, triturated in a Turratrec crusher and sifted to 60-mesh size particles, thus yielding the whole base flour. Part of the flour was defatted with petroleum ether p.a. (b.p. 30-60 °C) for protein evaluation (isolation, composition and digestibility).

2.2 - Methods

2.2.1 - Proximate analysis of kernels

For the proximate analysis, the moisture content was determined by drying samples in a stove at 105 °C according to the method described in the analytical norms of INSTITUTO ADOLFO LUTZ [16]. The total protein was determined

by the total nitrogen content (%) according to the Kjeldahl method described in AOAC [4] adopting a nitrogen conversion factor of 5.75, and also by using a procedure developed by BRADFORD [6], which uses bovine serum albumin (BSA) as the protein standard. Crude lipid (ether extract) and ash (fixed mineral residue) contents were analyzed according to AOAC [4]. Furthermore, dietary fiber was determined by the neutral detergent fiber method [32]. The total sugar content was determined by the reduction method according to the procedure described in the analytical norms of INSTITUTO ADOLFO LUTZ [16].

2.2.2 - Isolation of meal proteins

The seed protein fractions used in the present study — namely albumins, globulins, prolamins, glutelins and residue — were prepared according to a sequential extraction procedure with NaCl, ethanol and NaOH [19]. Fifteen-gram portions of kernel flour were extracted with 150 mL of 4% NaCl for 1 h. The slurry was centrifuged at 17000 x g for 30 min at 4 °C and the supernatant was then dialyzed against distilled water to separate the albumin and globulin. The residue of the salt extraction was suspended in 70% ethanol for 1 h and again centrifuged as described above to obtain prolamins. An alcohol-insoluble pellet was suspended in 0.1 M NaOH and extracted for 1 h. Glutelins were then obtained by centrifugation as described above. All the fractions plus the final insoluble residue were recovered by dialysis and freeze-drying.

2.2.3 - Molecular mass estimation

SDS-PAGE was performed according to LAEMMLI [17]. The molar mass of the proteins was calculated by comparing them with protein molar mass markers (phosphorylase b, 94 kDa; bovine serum albumin, 67 kDa; egg albumin, 43 kDa; carbonic anhydrase, 30 kDa; trypsin inhibitor, 20.1 kDa; lysozyme, 14.4 kDa).

2.2.4 - Amino acid composition

An amino acid analysis was performed on a PicoTag amino acid analyzer (Waters) as described by HENRIKSON & MEREDITH [12]. One nanomole of protein fraction was hydrolyzed in 6 M HCl/1% phenol at 106 °C for 24 h. The hydrolysate reacted with 20 µL of fresh derivatization solution (methanol: triethylamine: water: phenylisothiocyanate, 7:1:1:1, v/v) for 1 h at room temperature. After pre-column derivatization, phenylisothiocyanate (PTC) amino acids were identified on a reverse-phase HPLC-column by comparing their retention times to those of standard PTC amino acids (Pierce). Cysteine residues were quantified as a cysteic acid. Based on amino acid composition, the essential amino acid score according to HENLEY & KUSTER [11] was estimated, where:

$$\text{Score (\%)} = \frac{\text{the most limiting amino acid of the test protein}}{\text{the same most limiting amino acid in the reference pattern}} \times 100 \quad (1)$$

2.2.5 - Inhibitory activity assay

Bovine pancreatic trypsin and bovine pancreatic chymotrypsin were used for the enzymatic assays. Trypsin-like activities were assayed using N α -benzoyl-DL-arginine *p*-nitroanilide (BAPNA) as a substrate. Chymotrypsin-like activities were assayed using N-benzoyl-L-tyrosine *p*-nitroalide (BTpNA) as a substrate [20]. In a standard assay, a reaction mixture contained 50 μ L of each enzyme extract, a reaction buffer (0.1-M Tris-HCl buffer, pH 8.0) and 50 μ L of 1-mM substrate to a final volume of 500 μ L. The reaction was stopped by adding 200 μ L of 30% acetic acid. The release of *p*-nitroaniline groups was measured spectrophotometrically at 410 nm.

The protease inhibitor was assayed by preincubating 50 μ L of each fraction at concentrations ranging from 25 to 200 μ g with 50 μ L of protease and 350 μ L of the reaction buffer at 37 °C for 15 min. The reaction was started by adding the substrate and was done as described above. The remaining activity was expressed as the percentage of the enzymatic activity in the absence of the inhibitor.

2.2.6 - Hemagglutination assay

Hemagglutination assays were done on microtiter U-plates using serial dilutions with 50- μ L volumes of 0.15-M NaCl. A 50- μ L volume of a 2% suspension of type A human erythrocytes was added and after 1 h at room temperature, the results were read. The hemagglutination titer, defined as the reciprocal of the highest dilution showing hemagglutination, was defined as one hemagglutination unit [10].

2.2.7 - *In vitro* protein digestibility

This assay was carried out according to OSHODI *et al.* [24]. A 31.25 mg sample of protein (either globulin or glutelin) was dissolved in 5 mL of distilled water and the pH was adjusted to 8.0 with 0.1N HCl and/or 0.1N NaOH while stirring at 37 °C. A multienzyme solution consisting of 1.6 mg of trypsin, 3.1 mg of chymotrypsin and 1.3 mg of peptidase per mL was maintained in an ice bath and adjusted to pH 8.0 as described above. A 0.5 mL aliquot of the multienzyme solution was added to the protein sample solution and was constantly stirred at 37 °C. The pH of the solution was recorded 10 min after adding the enzyme solution. The *in vitro* digestibility was calculated using the HSU *et al.* equation [15].

$$Y = 210.46 - 18.1X \quad (2)$$

where Y is the *in vitro* digestibility (%) and X is the pH of the sample suspension after 10 min digestion with the multienzyme solution. A casein standard from Sigma was also analyzed as a reference for *in vitro* digestibility.

The relative molecular masses of the digestion products were estimated by SDS-PAGE using protein markers of a known molecular mass.

2.2.8 - Mineral analysis

The analysis was carried out according to the methodology described by SALINAS & GARCIA [28] involving organic digestion with acids. An atomic absorption spectrophotometer with acetylene (Perkin-Elmer 2380) was used to determine calcium, magnesium, iron, manganese, zinc and copper contents with wavelengths and slits of, respectively, 422.7 nm and 0.7 mm; 285.2 nm and 0.7 mm; 248.3 nm and 0.2 mm; 279.5 nm and 0.2 mm; 213.9 nm and 0.7 mm; and 324.7 nm and 0.7 mm. A flame photometer (Micronal B262) was used to determine the sodium (589 nm) and potassium (768 nm). A visible light spectrophotometer (Femto 482) was used for phosphorus (420 nm).

2.2.9 - Statistical analysis

The results were expressed as means \pm standard deviation, when applicable. Data underwent an analysis of variance (ANOVA) (general linear models, or GLM, procedure). Whenever a difference was found between treatments, the Student's *t*-test was used to determine the level of significance ($p < 0.05$) [26].

3 - RESULTS

3.1 - Proximate analysis

Table 1 shows the results of the proximate analysis of kernels. The kernels were found to have high protein content (17.6%), as compared to some *cerrado* fruit kernels and other palm tree species [3, 21]. Furthermore, they showed quite high lipid content (51.7%). Such protein and lipid values are significant, as they constitute, respectively, 12.5% and 82.9% of the total energy, showing that bocaiuva kernels are a good energy source. The kernels also have high fiber content (15.84%).

TABLE 1 – Proximate composition of kernels of bocaiuva, *Acrocomia aculeata* (Jacq.) Lodd., expressed as g/100 g of integral matter.

Component	Results*
Moisture	6.50 \pm 0.06
Ash	1.99 \pm 0.02
Crude lipid (ether extract)	51.71 \pm 2.90
Reducing sugars, in glucose	1.55 \pm 0.12
Nonreducing sugars, in sucrose	ND
Nonreducing sugars, in starch	4.84 \pm 0.25
Protein (Kjeldahl-N** x 5.75)	17.57 \pm 0.57
Total dietary fiber	17.23 \pm 0.49
Total calorie content (kcal/100 g)	561.23 \pm 26.04

ND = not detected; *Mean values \pm standard deviation of two triplicate determinations; and **Nitrogen by the Kjeldahl method.

3.2 - Isolation of kernel proteins

The protein content of the defatted kernel flour (as determined by Bradford's method) was 32.5%. The soluble proteins were fractionated according to their solubility. Most

of these proteins were present in the globulin (salt soluble) (53.5%) and glutelin (alkali soluble) (40.0%) fractions (Table 2). The extraction with organic solvent showed that the seeds were almost devoid of prolamins (1.1%).

TABLE 2 – Soluble protein fractions in kernels of bocaiuva, *Acrocomia aculeata* (Jacq.) Lodd.

Protein fraction	Protein (%) [*]
Globulin	53.5
Glutelin	40.0
Albumin	5.4
Prolamin	1.1

3.3 - Molecular mass estimation

As revealed by SDS-PAGE (Figure 1), the globulin fraction showed three protein bands, one of them above 100 kDa, one of 66 kDa and one of 60 kDa, all of which were absent from the glutelin fraction. However, other protein bands with the same profile were seen in both protein fractions. Lanes 3, 4 and 5 in Figure 1 also show that the globulin and glutelin fractions were digested by multienzymes. The glutelin fraction was less susceptible to digestion by enzymes than its globulin counterpart. Both fractions showed a protein band (14 kDa) that was not digested by enzyme complexes. This polypeptide was particularly resistant to attack by digestive enzymes. Protein bands above 22 kDa were more easily digested.

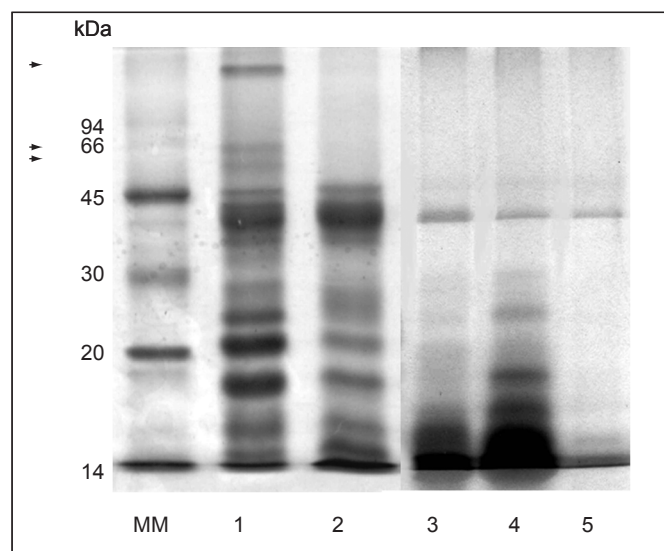


FIGURE 1 – SDS-PAGE of globulins and glutelins in kernels of bocaiuva, *Acrocomia aculeata* (Jacq.) Lodd. Lane MM, molecular mass markers; lane 1, globulin; lane 2, glutelin; lane 3, globulin digested with multienzyme solution; lane 4, glutelin digested with multienzyme solution; and lane 5, multienzyme solution.

3.4 - Amino acid composition

The amino acid profiles of the kernel protein fractions are shown in Table 3 beyond the proposed FAO/WHO [8] amino acid requirement pattern for the pre-school age group

(2-5 year old). Results show that bocaiuva protein fractions are potentially good sources of some essential amino acids, as they are rich in methionine + cysteine, valine and leucine. According to the FAO requirement standard, globulin, prolamins and albumin fractions of the bocaiuva kernel showed threonine as first limiting amino acid, revealing that it was also deficient in histidine, phenylalanine + tyrosine and lysine contents; and regarding the glutelin fraction, lysine was found to be the most limiting amino acid, followed by histidine and phenylalanine + tyrosine.

TABLE 3 – Amino acid composition of kernels of bocaiuva protein fractions, *Acrocomia aculeata* (Jacq.) Lodd., expressed as g/100 g protein.

Amino acid	Bocaiuva		Proteins		FAO ¹
	Globulin	Glutelin	Prolamin	Albumin	
Aspartic acid	6.25	8.70	6.87	7.27	-
Glutamic acid	20.43	15.65	22.85	21.83	-
Serine	5.24	6.09	4.27	4.23	-
Glycine	8.55	8.66	10.32	8.68	-
Histidine	1.34	1.57	0.87	1.14	1.9
Arginine	13.37	11.74	10.09	11.45	-
Threonine	1.60	3.63	1.40	1.69	3.4
Alanine	6.63	6.95	8.53	6.82	-
Proline	4.71	4.60	6.26	6.04	-
Valine	6.24	6.87	5.13	5.70	3.5
Methionine + Cysteine	3.15	3.87	3.63	4.20	2.5
Isoleucine	3.38	4.13	2.56	2.91	2.8
Leucine	7.19	6.92	6.80	7.09	6.6
Phenylalanine + Tyrosine	6.21	5.72	5.21	5.08	6.3
Lysine	5.72	2.87	5.19	5.75	5.8
Essential amino acid score (%)	47.1	49.5	41.2	49.7	-
Limiting amino acid	Thr	Lys	Thr	Thr	-

Note: Tryptophan was not determined. ¹ FAO/WHO requirement standard (1991) for the pre-school age group (2-5 year olds). Values found for bocaiuva kernel aminoacids are the average of two repetitions.

3.5 - Protease-inhibitory and hemagglutinating activities

The protease-inhibitory activity of the kernel proteins is summarized in Table 4. Neither globulins nor glutelins showed trypsin or chymotrypsin inhibitory activities. The kernel extracts agglutinated human erythrocytes at very high titers.

TABLE 4 – Protease-inhibitory and hemagglutinating activity of kernel proteins of bocaiuva, *Acrocomia aculeata* (Jacq.) Lodd.

Proteins	Hemagglutinating activity (titer) [*]	Proteases	
		Trypsin	Chymotrypsin
Globulin	2	ND	ND
Glutelin	3	ND	ND

^{*}Titer is defined as the reciprocal of the endpoint dilution that caused detectable agglutination of erythrocytes. The amount of proteins used in the assays was 500 µg/mL; and ND: not detected.

3.6 - *In vitro* protein digestibility

The digestion of native and heated globulins and glutelins by a multienzyme solution consisting of trypsin, chymotrypsin and peptidase was compared to that of a casein standard (Figure 2). The mean values for *in vitro* protein digestibility of kernel globulins, glutelins and caseins (for boiled and non-boiled samples) were respectively 78% and 82% for globulins, 69% and 72% for glutelins and 80% and 84% for caseins. Heat treatment did not significantly increase the enzymatic hydrolysis of the globulins and glutelins investigated. *In vitro* digestibility of native and heated kernel globulins was similar to that of casein. Under boiling and non-boiling conditions, the casein standard and the bocaiuva kernel globulins were more easily digested than the glutelins, whose proteases were investigated.

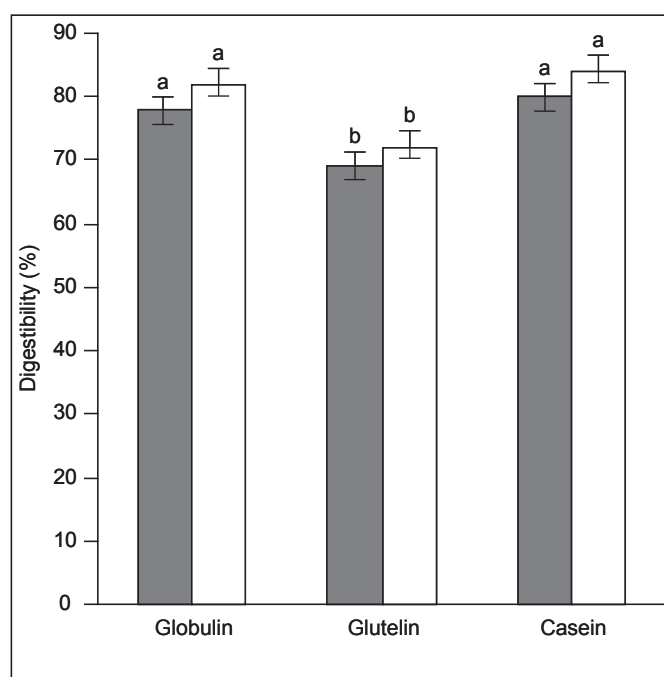


FIGURE 2 – *In vitro* digestibility of globulins and glutelins of bocaiuva, *Acrocomia aculeata* (Jacq.) Lodd., compared to a casein standard (mean \pm SD, n = 3). Dark columns are unboiled samples; white columns are boiled samples. Repeated letters mean that no significant differences were found among treatments, according to Student's *t* test (p < 0.05).

3.7 - Mineral contents

The mineral contents shown in Table 5 are high when compared to the values obtained for other palmtree fruits of Mato Grosso do Sul [13]. Bocaiuva kernels are richer in manganese, copper and zinc (respectively, 24.3, 11.1 and 30.9 $\mu\text{g/g}$) than other native fruits of the region; only kernels of piqui (*Caryocar brasiliense* Camb., a native tree of the *cerrado*) are described in the literature as richer in zinc and copper (respectively, 53.6 and 15.9 $\mu\text{g/g}$) than the bocaiuva kernels [13].

TABLE 5 – Mineral content of kernels of bocaiuva, *Acrocomia aculeata* (Jacq.) Lodd., expressed in a wet weight basis.

Mineral	mg/100 g
Calcium	94.3
Magnesium	207.0
Phosphorus	537.5
Potassium	377.2
	$\mu\text{g/g}$
Sodium	21.41
Iron	32.91
Manganese	24.33
Zinc	30.93
Copper	11.13

Values are the average of two repetitions.

4 - DISCUSSION

Separation and isolation based on solubility is a convenient system to begin the characterization of seed storage proteins from species that have not been studied yet [9].

In the present investigation, it was found that the defatted kernel flour of bocaiuva is a rich source of oil and protein. The kernel protein fraction is mostly composed of globulins (soluble in dilute saline solution) and glutelins (soluble in dilute acids or alkalis). Isolation of seed proteins has revealed that many plants (including Leguminosae) contain globulins as the main storage protein (50-75% of total seed protein) [5, 7, 23, 31]. Studies regarding the degradation of legume proteins, particularly globulins, have demonstrated that cooking and fermentation separates this protein fraction from antinutritional factors, such as proteinase inhibitors and lectins, which is a feature that makes globulin suitable for studies concerning the nutritional value of proteins [5, 7].

The agglutinating activity has been attributed to lectin (a hemagglutinating glycoprotein). In this study, the amount of it used in the agglutination assay was very high, indicating that bocaiuva globulin and glutelin fractions have very low lectin contents, in contrast with various plant seeds from which lectin has been isolated [7, 20, 29, 33].

As shown in Figure 2, a good level of *in vitro* digestibility was found for native and heated bocaiuva kernel globulins, as compared to the casein standard. However, the glutelin sample evaluated was less easily digested by proteases than casein, both under boiling and non-boiling conditions. The digestibility of bocaiuva globulin was similar to that of karkade flour (82.14%) [1]. Results for bocaiuva differed from those obtained elsewhere for proteins from legume seeds, such as soy, algaroba (mesquite) and cowpea, which are resistant to hydrolysis by mammalian digestive enzymes, given the presence of protease inhibitors in the seeds [5, 24, 31].

The digestibility of the glutelin fraction of bocaiuva was also lower than that reported in the literature for the karkade protein [1] though similar to that of legume seed proteins, such as the chickpea and jojoba [23, 31].

The nonsignificant increase in the digestibility of bocaiuva kernel globulins and glutelins after boiling (Figure 2) revealed that neither protein fraction contains protease inhibitors associated with low protein digestibility of plant seeds [5, 33, 31].

When the mineral profiles shown in Table 5 are compared to those of selected edible seeds, bocaiuva kernels are found to be richer in calcium (94.3 mg/100 g), phosphorus (537.5 mg/100 g) and manganese (24.3 µg/g) than cashew nuts or fresh coconut [25]. The values found for minerals in the samples investigated, however, were below the recommended dietary allowances for adults [22].

The nutritional value of dietary proteins is determined by the profile and quantity of the essential amino acids they contain. The presence of one or more essential amino acids in adequate amounts increases the nutritional value of a protein. The amino acid profile (Table 3) shows that bocaiuva kernel protein fractions are potentially good sources of some of the essential amino acids as they are rich in methionine + cysteine, valine and leucine. Threonine and lysine are the most limiting amino acids, respectively, from the two main protein fractions of bocaiuva kernel, globulin (amino acid score of the 47.1%) and glutelin (amino acid score of the 49.5%), in terms of the theoretical profiles for children in the age range of 2 to 5 years old. Methionine is an amino acid found at high concentrations in the proteins of other Brazilian fruits, such as castanha do Pará [27] and anajá [3] with values of 7.61% and 2.62% of the total protein content, respectively. The glutelin fraction of bocaiuva kernels have high nutritional values of methionine (3.3% of the glutelin fraction), if compared to the FAO's reference standards [8]. Plant storage proteins are generally rich in proline, asparagine, glutamine and arginine [7]. In bocaiuva kernel protein fractions, the predominant amino acids, in relation to the total found, were glutamine and arginine. In the globulin fraction (the main protein fraction of bocaiuva), the arginine and glutamine contents were found to be compatible with those determined by AMAYA-FARFÁN *et al.* [3] for another species of the same genus, *A. sclerocarpa* Mart. (13.86% and 19.79%, respectively). In comparison to fruit from the cerrado, the jatobá-do-cerrado (*Hymenaea stigonocarpa* Mart.) seed showed that protein was deficient in sulphur amino acids and threonine and rich in other essential amino acids [21].

5 - CONCLUSIONS

Bocaiuva kernels are a valuable source of total proteins and lipids, and in contrast with legume seeds, do not contain protease inhibitors — a nutritional advantage in terms of digestibility. The kernels, however, are limiting in some essential amino acids such as threonine and lysine (the main limiting amino acids), a feature that suggests the need for additional studies to provide a better understanding of the *in vivo* protein digestibility and biological value of bocaiuva kernels. As for the mineral analysis, the samples analyzed showed lower values than those recommended as dietary allowances for adults.

6 - REFERENCES

- [1] ABU-TAR BOUSH, H. M.; AHMED, S. A. B. Studies on karkade (*Hibiscus sbadariffa*): protease inhibitor, phytate, *in vitro* protein digestibility and gossypol content. **Food Chemistry**, v. 56, n. 1, p. 15-19, 1996.
- [2] ALMEIDA, S. P. **Cerrado-aproveitamento alimentar**. Planaltina: EMBRAPA-CPAC, 1998. 188 p.
- [3] AMAYA-FARFÁN, J.; RODRIGUEZ-AMAYA, D. B.; NOLETO CRUZ, P.; MARQUES, E. P. Fatty acid and amino acid composition of some indigenous fruits of northeastern Brazil. **Ciência e Tecnologia de Alimentos**, v. 6, n. 1, p. 86-92, 1986.
- [4] AOAC. **Official methods of analysis**, 16th ed. Washington DC, USA: Association of Official Analytical Chemists, 1995. 109 p.
- [5] ARAUJO, A. H.; CARDOSO, P. C. B.; PEREIRA, R. A.; LIMA, L. M.; OLIVEIRA, A. S.; MIRANDA, M. R. A.; XAVIER-FILHO, J. SALLES, M. P. *In vitro* digestibility of globulins from cowpea (*Vigna unguiculata*) and xerophytic algaroba (*Prosopis juliflora*) seeds by mammalian digestive proteinases: a comparative study. **Food Chemistry**, v. 78, p. 143-147, 2002.
- [6] BRADFORD, M. M. A rapid and sensitive method for the quantification of microgram quantities of protein using the principle of protein-dye binding. **Analytical Biochemistry**, v. 72, p. 248-254, 1976.
- [7] DERBYSHIRE, E.; WRIGHT, D. J.; BOUTER, D. Legumin and vicilin storage proteins of legume seeds. **Phytochemistry**, v. 15, p. 3-24, 1976.
- [8] FAO/WHO. **Protein quality evaluation**. Rome, Italy: Food and Agricultural Organization of the United Nations, 1991.
- [9] FONSECA, P. A.; FERREIRA, R. B.; TEIXEIRA, A. R. Seed proteins from *Quercus suber*. **Journal of Agricultural and Food Chemistry**, v. 45, p. 3443-3447, 1997.
- [10] FREIRE, M. G. M.; GOMES, V. M.; CORSINI, R. E.; MACEDO, M. L. R. Isolation and partial characterization of a novel lectin from *Talisia esculenta* seeds that interferes with fungal growth. **Plant Physiology and Biochemistry**, v. 40, p. 61-68, 2002.
- [11] HENLEY, E. C.; KUSTER, J. M. Protein quality evaluation by protein digestibility – corrected amino acid scoring. **Food Technology**, v. 48, p. 74-77, 1994.
- [12] HENRIKSON, R. L.; MEREDITH, S. C. Amino acid analysis by reverse phase high performance liquid chromatography: precolumn derivatization with phenylisothiocyanate. **Analytical Biochemistry**, v. 36, p. 65-71, 1984.
- [13] HIANE, P. A.; RAMOS, M. I. L.; RAMOS FILHO, M. M.; BARROCAS, G. E. G. Teores de minerais de alguns frutos do Estado de Mato Grosso do Sul. **Boletim do Centro de Pesquisa e Processamento de Alimentos**, v. 10, n. 2, p. 135-150, 1992a.
- [14] HIANE, P. A.; RAMOS, M. I. L.; RAMOS FILHO, M. M.; PEREIRA, J. G. Composição centesimal e perfil de ácidos graxos de alguns frutos nativos do Estado de Mato Grosso do Sul. **Boletim do Centro de Pesquisa e Processamento de Alimentos**, v. 10, n. 1, p. 35-42, 1992b.

- [15] HSU, H. W.; VAVAK, D. L.; SATTERLEE, L. D.; MILLER, G. A. A multienzyme technique for estimating protein digestibility. **Journal of Food Science**, v. 42, p. 1269-1273, 1977.
- [16] INSTITUTO ADOLFO LUTZ. **Normas analíticas do Instituto Adolfo Lutz**. 3rd ed. São Paulo, 1985. 533 p.
- [17] LAEMMLI, U. K. Cleavage of structural proteins during the assembly of the head of bacteriophage T4. **Nature**, v. 227, p. 680-685, 1970.
- [18] LIENER, I. E. Implications of antinutritional components in soybean foods. **Critical Review of Food Science and Nutrition**, v. 34, p. 31-67, 1994.
- [19] MACEDO, M. L. R.; DAMICO, D. C. S. Effects of protein fractions from *Zea mays* L. on development and survival of mexican bean weevil *Zabrotes subfasciatus* (Boh.). **Insect Science and Its Application**. v. 20, p. 135-139, 2000.
- [20] MACEDO, M. L. R.; FREIRE, M. G. M.; CABRINI, E. C.; TOYAMA, M. H.; NOVELLO, J. C.; MARANGONI, S.; MATOS, D. G. G. A trypsin inhibitor from *Peltophorum dubium* seeds active against pest proteases and its effects on the survival of *Anagasta kuehniella* (Lepidoptera: Pyralidae). **Biochimica et Biophysica Acta**, v. 1621, p. 170-182, 2003.
- [21] MATUDA, T. G.; MARIA NETTO, F. Caracterização química parcial da semente de jatobá-do-cerrado (*Hymenaea stigonocarpa* Mart.). **Ciência e Tecnologia de Alimentos**, v. 25, n. 2, p. 353-357, 2005.
- [22] NATIONAL RESEARCH COUNCIL. **Recommended dietary allowances**, 10th ed. Washington, D. C.: National Academy of Sciences, 1989.
- [23] NEVES, V. A.; SILVA, M. A.; LOURENÇO, E. J. Caracterização e hidrólise da globulina principal de grão de bico (*Cicer arietinum* L.), var. IAC - Marrocos. **Ciência e Tecnologia de Alimentos**, v. 24, n. 1, p. 139-145, 2004.
- [24] OSHODI, A. A.; IPINMOROTI, K. O.; ADEYEYE, E.I.; HALL, G.M. *In vitro* multienzyme digestibility of protein of six varieties of African yam bean flours. **Journal of Science and Food Agricultural**, v. 69, p. 373-377, 1995.
- [25] PHILIPPI, S. T. **Tabela de composição de alimentos: suporte para decisão nutricional**. 2nd ed. São Paulo: Coronário, 2002. 135 p.
- [26] PIMENTEL GOMES, F. **Curso de estatística experimental**. 14th ed. Piracicaba: F. Pimentel-Gomes Ed. 2000. 477 p.
- [27] RIBEIRO, M. A. A. **Aproveitamento tecnológico de castanha-do-Brasil (*Bertholletia excelsa*): estudo da conservação**. Dissertação (Mestrado). Piracicaba: Escola Superior de Agricultura "Luis de Queiroz"/USP, 1982. 88 p.
- [28] SALINAS, Y. G.; GARCIA, R. **Métodos químicos para el análisis de suelos ácidos y plantas forrajeras**. Cali: Centro Internacional de Agricultura Tropical, 1985. 83 p.
- [29] SEENA, S.; SRIDHAR, K. R. Nutrient composition and biological evaluation of an unconventional legume, *Canavalia cathartica* of mangroves. **International Journal of Food Science and Nutrition**, v. 55, p. 615-625, 2004.
- [30] SGARBIERI, V. C. **Proteínas em alimentos protéicos: propriedades - degradações - modificações**. São Paulo: Varela, 1996. 517 p.
- [31] SHRESTHA, M. K.; PERI, I.; SMIRNOFF, P.; BIRK, Y.; GOLAN-GOLDHIRSH, A. Jojoba seed meal proteins associated with proteolytic and protease inhibitory activities. **Journal of Agricultural and Food Chemistry**, v. 50, p. 5670-5675, 2002.
- [32] VAN SOEST, P. J.; WINE, R. H. Use of detergents in the analysis of fibrous food. IV. Determination of cell-wall constituents. **Journal of Association of Analytical Chemistry**, v. 50, p. 50-55, 1967.
- [33] VASCONCELOS, I. M.; MAIA, A. A. B.; SIEBRA, E. A., OLIVEIRA, J. T. A.; CARVALHO, A. F. F. U.; MELO, V. M. M.; CARLINI, C. R.; CASTELAR, L. I. M. Nutritional study of two Brazilian soybean (*Glycine max*) cultivars differing in the contents of antinutritional and toxic proteins. **Journal of Nutrition and Biochemistry**, v. 12, p. 55-62, 2001.

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