Journal of Applied Botany and Food Quality 83, 151 - 156 (2010)

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Nutritional, bioactive and enzymatic properties of *Ouratea fieldingiana* (Gardner) Engl. (Ochnaceae) fruits. A little-known plant from Northeastern Brazil

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(Received October 16, 2009)

Summary

The species *Ouratea fieldingiana* is a bush belonging to the family Ochnaceae, occurring frequently in the "Caatinga" Dominium. This species and other members of the genus *Ouratea* are rarely studied and no data are available on the chemical composition or medical and industrial potential of this plant. Thus, this work aimed to study the nutritional, medical and industrial potential of *O. fieldingiana* fruits by determining the proximate composition, caloric value and the content of toxic and antinutritional factors. The search for biological and enzymatic activities with pharmacological and industrial interest was also conducted. The results showed that the dry fruits are rich in proteins (21.58 ± 2.02 g/100g) and dietary fiber (46.72 ± 0.52 g/100g), do not show trypsin inhibitors, toxins or tannins, and have low levels of urease (2.41 ± 0.22 units kg^{-1}) and lectins (780 and 3,098 HU/g protein in native erythrocytes and in protease-treated erythrocytes, respectively). The fruit aqueous extracts showed antibacterial activity (growth inhibition halo with diameter > 10 mm) against *Cromobacterium violaceum*, possibly due to a protein compound, and also amylolytic (starch degradation area > 60 mm^2), hemolytic (100% hemolysis in rat and mouse erythrocytes') and strong anti-coagulant (persisted for more than 24 h) activities. The *O. fieldingiana* fruits are potentially nutritious, showing high protein and dietary fiber levels, as well as a low content of toxic and/or antinutritional compounds. The fruits aqueous extracts possess bioactive compounds presenting pharmacological and/or industrial interest.

Introduction

"Caatinga" forest is a biome from Northeastern Brazil known for presenting a high biodiversity, although it is quite scientifically underexploited. Despite the lack of research dedicated to "Caatinga", there is a growing use of its native vegetation for profit purposes only. Thus, basic sustainability elements are required to prevent extinction of native species, since potential properties and applications are still unknown (ARAÚJO FILHO and BARBOSA, 2000). Among several plant species in "Caatinga", *Ouratea fieldingiana* was chosen as object of study in this work due to lack of information on its chemical composition and the presence of interesting industrial and/or pharmacological bioactive compounds. This species belongs to the Ochnaceae family, Theales order (DAHLGREN, 1980), which comprises about 30 genera and 400 species widely distributed in tropical and subtropical regions around the world. In Brazil, there are approximately 10 genera with 105 species (JOLY, 1988). They are essentially trees or shrubs with fruits which are small drupes arranged in clusters. Each drupe contains only one ovoid seed (BRAGA, 1976). All the species of the *Ouratea* genus are known in Northeastern Brazil as "Batiputá" and recognized as rich in biflavonoids (DANIEL et al., 2005; FERREIRA et al., 2006). Some members of the *Ouratea* genus are widely used in folk medicine to treat dysentery, diarrhea, rheumatism and gastric diseases (BOUQUET, 1969).

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This work intended to study the nutritional, bioactive and enzymatic properties of pharmacological and/or industrial interest of *O. fieldingiana* fruits, with the major goal of encouraging research on this species in several areas.

Materials and methods

Plant material and processing

O. fieldingiana fruits were collected in Crato (Ceará State, Brazil) in January 2005 and identified by Edson de Paula Nunes, at Prisco Bezerra Herbarium, Federal University of Ceará. Some specimens were incorporated at the herbarium of the same institution under the identification number EAC 34710. Fruits were ground in a coffee mill and exposed to 47 °C for 72 h. Conventional sieves of approximately 1 mm^2 mesh were used to obtain a homogeneous powder, which was protected from excessive moisture and wrapped in plastic containers for use in subsequent trials.

Crude aqueous extract preparation

Powdered material from *O. fieldingiana* dehydrated fruits was mixed with 50 mM sodium borate buffer, pH 8.0, and 150 mM NaCl 1:5 (w/v) and left under continuous stirring for 2 h at 4 °C. Suspension was filtered and then centrifuged at 20,000 g for 20 min at 4 °C. The precipitate was submitted to a further extraction, under the same conditions described above. The supernatants obtained from both extractions were mixed and used in the subsequent tests. In order to predict molecular mass magnitude and thermostability of active(s) principle(s) in crude extract (CE), the dialysed crude extract (DCE) and heated crude extract (HCE) were prepared. DCE was obtained from exhaustive dialysis of CE against distilled water in a 6-8 kDa cutt-off dialysis membrane (Fisherbrand, Pittsburgh, USA). HCE was obtained by heating the CE at 100 °C for 30 min and then centrifuged at 20,000 g for 20 min, being the precipitate discarded and the supernatant used in the biological assays. All determinations and assays described below were run with at least triplicates.

Soluble proteins and carbohydrates content and total soluble solids determination

Soluble proteins content in CE, DCE and HCE was estimated by the colorimetric method of Coomassie Brilliant Blue according to BRADFORD (1976) and soluble carbohydrate content according to DUBOIS et al. (1956), using bovin serum albumin and glucose as standards, respectively. The total soluble solids (TSS) content was estimated by measuring the residual mass (g) into 1.0 mL of the extracts after evaporation in oven at 100 °C for 24 h.

Proximate composition and energy

Total protein analysis followed the manual colorimetric procedure for measuring ammonium nitrogen (BAETHGEN and ALLEY, 1989).

The other constituents of proximate composition, lipid, mineral matter, starch and total sugars (reducing and non-reducing sugars) were determined according to AOAC (1997). The total dietary fiber content was measured by the enzyme-gravimetric method (AOAC, 1997) and digestible carbohydrate was determined by percentage difference of all other constituents. The results were expressed in g/100 g meal of dehydrated fruits. The dried fruit energy was calculated by multiplying the crude protein and digestible carbohydrates percentages sum by the factor 4 (kcal g^{-1}) and added total lipid content multiplied by the factor 9 (kcal g^{-1}).

Toxic and/or antinutritional factors

Trypsin inhibitors content was determined according to the methodology proposed by KAKADE et al. (1974). The agglutinins (lectins) were detected by methodology described by MOREIRA and PERRONE (1977), using rabbit erythrocytes, treated and non-treated with *Bacillus licheniformis* proteases (Sigma-Aldrich Co. USA). Urease content was measured using methodology proposed by KAPLAN (1969). To assess the presence of toxins, an acute toxicity test in mice (n = 6) was run by intraperitonial injection (30 mL/kg body weight) of CE according to VASCONCELOS et al. (1994). All procedures were approved by the Animal Experimentation Ethics Committee of Universidade Federal do Ceará (CEPA) which adopts the guidelines of Brazilian College of Animal Experimentation (COBEA).

Antimicrobial and larvicidal activities against *Aedes aegypti*

Antibacterial activity of *O. fieldingiana* CE, DCE and HCE was determined as described by BAUER et al. (1966). The Gram-positive bacteria *Staphylococcus aureus* (ATCC 25923), *Bacillus subtilis* (ATCC 6633) and *Cromobacterium violaceum* (ATCC 12472) and the Gram-negative bacteria *Enterobacter aerogens* (ATCC 13048), *Salmonella choleraesuis* (ATCC 10708), *Klebsiella pneumoniae* (ATCC 10031) and *Pseudomonas aeruginosa* (ATCC 25619) were obtained from American Type Culture Collection (ATCC). Paper filter discs with 20 µL of CE, DCE and HCE were placed on Mueller-Hinton agar sterile plates containing bacteria cultures at a concentration of 10^7 CFU mL^{-1}. Plates were incubated at 37 °C for 24 h. Tetracycline was used as positive control and 50 mM sodium borate buffer, pH 8.0 with 150 mM NaCl as negative control. Microbial growth inhibition was assessed by formation of inhibition halos around the discs.

The test for antifungal activity was performed according to the method described by ROBERTS and SELITRENNIKOFF (1990), with modifications. The fungi *Aspergillus fumigatus, A. niger, Colletotrichum gloesporioides, C. musae, C. truncatum, Fusarium oxysporum, F. solani, F. pallidoroseum, Mucor sp., Neurospora sp., Penicillium herguei, Phomopsis sp., Phytium oligandrum, Trichoderma viride* and *Rhizoctonia solani* were obtained from the Laboratory of Microbial Ecology and Biotechnology, Federal University of Ceará. Paper filter discs (2 cm diameter) with 300 µL of CE were placed on sterile Petri dishes containing potato-agar and a pellet of fungi mycelium. The plates were incubated in growth chambers with a photoperiod of 12 h until fungal growth. Halos formation around the discs indicated fungal growth inhibition. Nystatin (100,000 UI mL^{-1}, EMS, São Paulo) was used as positive control.

Ae. aegypti larvae were collected from a mosquito colony kept at NUVET (Núcleo de Controle das Endemias Transmissíveis por Vetores, Secretaria de Saúde do Estado do Ceará). The test was conducted according to the methodology described by FARIAS et al. (2009). Twenty *Ae. aegypti* larvae at 3rd instar were transferred to disposable plastic cups of 150 mL capacity containing 25 mL of *O. fieldingiana* CE prepared in distilled water. The test was conducted at room temperature (25 °C) and photoperiod of 12 h. Mortality and survival rates were recorded after 24 h. The tests were performed in triplicate and temephos (Sigma-Aldrich Co., USA) was used as positive control and distilled water as negative control.

Chitinase and peroxidase activities

Chitinase activity determination in the CE was carried out following the methodology described by BOLLER (1993), which measures N-acetyl-D-glucosamine (NAG) release due to hydrolytic enzymes action on colloidal chitin. *Serratia marcescens* chitinase (EC 3.2.1.14, Sigma-Aldrich Co. USA) was used as positive control. Peroxidase activity was evaluated in a SDS-PAGE according to LAEMMLI (1970), followed by the methodology described by CARUSO et al. (1999), with some modifications. The gel was immersed in 2.5% Triton X-100 solution for 30 min, at 25 °C then immersed in 50 mM sodium acetate buffer, pH 5.2. To reveal the peroxidase activity, the gel was dipped in a revealing solution, composed of 0.2% guaiacol (v/v), 0.01% 3-amino-9-ethyl-carbazole (w/v) and 0.03% peroxide hydrogen (v/v). The reaction was stopped by successive washing with distilled water. Peroxidase isoenzymes from Horseradish (Sigma-Aldrich Co. USA) were used as positive control.

Amylolytic, cellulolytic and proteolytic activities

Amylolytic activity was assessed according to HANKIN and ANAGNOSTAKIS (1975), with some modifications. Thirty microliters of each sample (CE, DCE and HCE) and positive control (60 IU mL^{-1} of α-amylase EC 3.2.1.1 from *Aspergillus oryzae*, Sigma-Aldrich Co. USA) were inoculated in wells drilled in agar-starch gel (10 g L^{-1}). After incubation at 37 °C for 24 h, plates were stained with lugol solution to allow visualization of substrate degradation by the formation of clear halos around the wells. Cellulolytic activity was determined according to the methodology described by RÜEGGER and TAUK-TORNISIELO (2004), with some modifications, in which lyophilized *O. fieldingiana* CE was dissolved in 50 µL (1:1, w/v) 50 mM sodium phosphate buffer, pH 6.0 containing NaCl 0.5 M, then inoculated in Petri dishes containing agar (20 g L^{-1}) and cellulose (10 g L^{-1}). *Aspergillus* sp. cellulase (EC 3.2.1.4., Sigma-Aldrich Co. USA) was used as positive control. We used Congo red dye solution (2.5 g L^{-1} in 0.1 M Tris HCl, pH 8.0) for 30 min, followed by washing with 5 mL 0.5 M NaCl solution in the same buffer to reveal cellulolytic activity. Formation of clear halos around the wells indicated cellulose degradation.

The method described by MICHAUD et al. (1996) was used for proteolytic activity detection in the CE sample, as follows. Samples were applied in a gel electrophoresis (SDS-PAGE) containing gelatin at 0.1%. The gel was then immersed in 2.5% Triton X-100 solution for 30 min, at 25 °C, and then immersed in the activator solution (25 mM sodium phosphate buffer, pH 6.0, containing 0.2 mM DTT and 1 mM EDTA) for 1 h at 37 °C. Bands with proteolytic activity were visualized by negative staining with the Coomassie Brilliant Blue R dye. Proteases from *Aspergillus oryzae* (EC 232-642-4, Sigma-Aldrich Co. USA) in a concentration of 1 mg mL^{-1} was used as positive control.

Hemolytic, anticoagulant and heparinase activities

Hemolytic activity in the CE, DCE and HCE samples was determined according to BERNHEIMER (1988), using native erythrocytes of rats and mice (1% suspension prepared in 0.15 M NaCl). Distilled water was used as positive control. Anticoagulant and heparinase activities in these extracts were performed according to methodology described by RAJAGANAPATHI and KATHIRESAN (2002), using freshly collected

blood of rats and mice. Sodic heparin 5,000 IU mL^{-1} (Liquemine®, Roche, Brazil) was used as positive control for anticoagulant assay.

Results and discussion

The results on the proximate composition and caloric value of *O. fieldingiana* fruits, on a dry basis, are described in Tab. 1. The *O. fieldingiana* fruit morphology, with a small layer of pulp and peel (mesocarp and exocarp, respectively) and a large seed has led to comparisons of the composition data with those of other seeds with significance as food source. Due to economic importance, nutritional quality and large quantity of studies, nutritional parameters of leguminous seeds was used in the discussion for comparative purposes. The fruits of *O. fieldingiana* were shown to be especially rich in protein (21.58 ± 2.02 g/100 g) and dietary fiber (46.72 ± 0.52 g/100 g). The high protein content is equivalent to those described for cowpea (*Vigna unguiculata* L. Walp), an important protein source for Northeastern Brazil population, which ranges from 19.5 to 26.1 g/100 g (MAIA et al., 2000). Although legumes are rich in proteins, they are low in essential sulfur amino acids (RAJARAM and JANARDHANAN, 1991). Thus, studies on the amino acids composition of *O. fieldingiana* fruits are also important to assess the protein quality. The total dietary fiber content was higher than that described by MECHI et al. (2005) in *Phaseolus vulgaris* bean (38.6 ± 1.6 g/100 g), which is one of the most important sources of dietary fiber for many people, and also similar to those described for species of the genus *Hymenaea* (53.87 to 55.6 g/100 g), an underexploited legume highly valued for its fiber content (SILVA et al., 2001). Although lipid fraction (12.57 ± 0.01 g/100 g) was much higher than that described for *P. vulgaris* (0.44 g/100 g), it was lower than that reported by VASCONCELOS et al. (2006) for different cultivars of soybean (20.06 ± 0.12 to 23.2 ± 0.47 g/100 g). The mineral matter content observed in *O. fieldingiana* (1.43 ± 0.04 g/100 g) was about four times lower than that reported for *G. max* (5.22 ± 0.05 g/100 g) by VASCONCELOS et al. (2006) and about three times lower than that of *P. vulgaris* (4.88 ± 0.01 g/100 g) (MECHI et al., 2005). The digestible carbohydrates content represent a small fraction of the composition of *O. fieldingiana* fruits, only 17.7 g/100 g, being slightly lower than that described for *P. vulgaris* (29.45 g/100 g) by MECHI et al. (2005). *O. fieldingiana* fruits are good sources of energy (270 kcal/100 g), while *P. vulgaris* beans contains 228.16 kcal/100 g (MECHI et al., 2005).

Data on the contents of toxic and/or antinutritional factors for *O. fieldingiana* fruits are shown in Tab. 2. These fruits were potentially nutritive, with low content of toxic and/or antinutritional factors.

Tab. 1: Proximate composition and caloric value of *Ouratea fieldingiana* fruits, in dry basis

Parameters analyzed	*O. fieldingiana* Fruits
Proximal composition (g/100 g)	
Total proteins[a]	21.58 ± 2.02
Total lipids	12.57 ± 0.01
Minerals	1.43 ± 0.04
Dietary fiber	46.72 ± 0.52
Digestible carbohydrates[b]	17.7
Energy[c] (Kcal./100 g)	270

Values are mean ± standard deviation of three analyses, each in triplicate;
[a] N total x 6.25;
[b] Calculated by the percentage difference of all other constituents;
[c] g of carbohydrate x 4 kcal + g of protein x 4 kcal + g of lipid x 9 kcal (MAHAN and SCOTT-STUMP, 1996).

Tab. 2: Toxic and/or antinutritional factors of *Ouratea fieldingiana* fruits

Toxic and/or antinutritional factors	*O. fieldingiana* fruits
Trypsin inhibitor	ND[a]
Lectin[b]	780[b1]
	3098[b2]
Urease[c]	2.41 ± 0.22
Toxin	NL[d]
Tannin	ND

Values are mean of three analyses, each in triplicate;
[a] Non-detected with the methodology used;
[b] The hemagglutinating activity is expressed in Units of Hemagglutination (UH) per g of protein;
[b1] Hemagglutinating activity in rabbit non-treated erythrocytes;
[b2] Hemagglutinating activity in rabbit erythrocytes, treated with protease;
[c] Urease activity is expressed in units of enzyme per kg of meal. The units were calculated using information from Sigma in which 1 g of pure enzyme contains 870,000 units;
[d] Not lethal at a dose of 1g/kg mice body weight.

Tannins, trypsin inhibitors and toxins were not detected. These results add positive attributes favoring their use, considering the fact that the main plant sources of protein and fiber (beans and soybeans) contain these and other toxic and/or antinutritional factors in much higher quantities. ENE-OBONG (1995) detected 1.42 mg tannin g $meal^{-1}$ in *V. unguiculata* seeds. Also, in different soybean cultivars, VASCONCELOS et al. (2001) reported trypsin inhibitors in the order of 30.6 ± 1.1 to 62.5 ± 2.6 g of inhibited trypsin kg^{-1} of meal and acute toxicity in mice with LD_{50} of 0.137 ± 0.02 g kg^{-1} body weight. The urease content described in *O. fieldingiana* fruits (2,410 ± 220 units of urease kg^{-1} meal) was very low compared to those described by VASCONCELOS et al. (2001) in soybeans (107,300 ± 9,500 to 219,300 ± 12,600). The hemagglutinating activity was calculated based on the specific activity of a lectin, according to the amount of proteins present in the extracts. The specific hemagglutinating activity of *O. fieldingiana* CE (non-treated and treated erythrocytes – 780 and 3,098 UH g^{-1} protein, respectively) was much lower than that described by VASCONCELOS et al. (2006) in several soybean cultivars (2,920 to 4,540, and 82,800 to 181,400 UH g^{-1} protein, respectively). The vast majority of known lectins are inactivated by cooking or roasting. Therefore the presence of these antinutritional factors in *O. fieldingiana* fruits does not represent a major problem to the use of this underexploited plant as food.

The results on the preliminary characterization of *O. fieldingiana* fruits aqueous extract (CE, DCE and HCE) concerning enzymatic and biological activities are shown in Tab. 3. After preliminary screening (data not shown), 50 mM sodium borate, pH 8.0, containing 150 mM NaCl was the most efficient buffer solution for extracting soluble proteins from *O. fieldingiana* fruits powder. The TSS of *O. fieldingiana* CE (33.95 ± 0.07 mg mL^{-1}) is constituted by a high content of carbohydrates (14.40 ± 0.11 mg mL^{-1}), but a low amount of proteins (4.12 ± 0.06 mg mL^{-1}). As expected, this profile was similar to HCE. Nevertheless, the HCE proteins probably had its biological properties severely compromised by high temperature and time exposure. The DCE showed lower amounts of carbohydrates and proteins and consequently of TSS due to dilution of the sample during dialysis. However, the protein content increased compared to amount of carbohydrates and TSS. These treatments given to CE may provide relevant clues about the molecular mass magnitude and thermostability of active principles. PEGNYEMB et al. (2005) reported antibacterial activity of biflavonoids extracted from *O. sulcata* against Gram-positive strains (*Staphyloccocus aureus* and *Bacillus subtilis*) and also against Gram-negative bacteria *Vibrium*

Tab. 3: Preliminary characterization of *Ouratea fieldingiana* fruit extracts (Crude Aqueous Extract – CE, Heated CE Extract – HCE and Dialysed CE Extract – DCE) and its enzymatic and biological activities

Parameters	Extracts of *O. fieldingiana* frutis		
	Crude aqueous extract (CE)	Heated CE (HCE)	Dialysed CE (DCE)
Constituents (mg · mL^{-1})[a]			
Soluble proteins	4.12 ± 0.06	3.50 ± 0.12	1.96 ± 0.06
Soluble carbohydrates	14.40 ± 0.11	10.44 ± 0.39	4.71 ± 0.04
Total soluble solids	33.95 ± 0.07	34.05 ± 0.92	9.41 ± 0.07
Presence (+) or absence (-) of activity			
Larvicidal against *Ae. aegypti*[b]	-	NA[c]	NA
Antibacterial[d]	+	-	+
Antifungal	-	NA	NA
Proteolytic	-	NA	NA
Chitinolytic	-	NA	NA
Peroxidasic	-	NA	NA
Cellulolytic	-	NA	NA
Amylolytic[e]	+	-	+
Hemolytic[f]	+	+	+
Heparinasic	-	NA	NA
Anticoagulant[g]	+	+	+

(+) = Detectecd;
(-) = Not-detected;
[a] Values are mean ± standard deviation of three analyses, each in triplicate;
[b] All activities was performed in triplicates and the result considered positive when similar to positive control used;
[c] NA = Not Achieved. After the verification of a negative result for CE the other extracts (HCE and DCE) were not investigated any further.
[d] Growth inhibition halo with diameter > 10 mm was considered positive result;
[e] Starch degradation halo with area > 60 mm^2 was considered positive result;
[f] % of hemolysis similar to positive control (distilled water) at 540 nm;
[g] Coagulation inhibition > 10 min was considered positive result.

anguillarum. *O. fieldingiana* CE showed activity against the Gram-positive strain *Cromobacterium violaceum*. However, it seems that this antibacterial activity is probably due to a protein, since *O. fieldingiana* DCE showed the most evident growth inhibition of *C. violaceum* (halo diameter >10 mm), whereas the CE a discrete action, and HCE showed no activity. The purification of this new protein with antibacterial properties and the understanding of its mechanism of action against *C. violaceum* will add data to the vast literature describing the antibacterial activity of peptides and proteins of plant origin (NG, 2004). Despite the widely reported activity of many plants extract against fungi (WEBSTER et al., 2008) and larvae of *Ae. aegypti* (CARVALHO et al., 2003; FERREIRA et al., 2009), these activities were not detected in the *O. fieldingiana* CE.

As seen in Tab. 3, the *O. fieldingiana* fruits aqueous extract showed no chitinase, peroxidase, protease and cellulase activity, but it did show amylolytic activity. The lack of chitinase activity was consistent with the results on antifungal and larvicidal activity. Chitin is a structural component of fungi cell walls and insect exoskeleton, so its degradation by chitinase works as a defense strategy for many plants, expressing this enzyme in their organs (YE and NG, 2005). Peroxidases are quite common in plants and are especially involved in defense against pathogens (BRADLEY et al., 1992). However, peroxidase activity was not detected in the *O. fieldingiana* CE. The absence of this activity may be due to the use of a different defense arsenal or because this protein expression is dependent on specific external signaling. Although proteases are usually part of the enzyme repertoire of natural plants due to either endogenous compounds expression or the endophytic microorganisms (CARRIM et al., 2006), proteolytic activity was not detected in CE. The presence of cellulolytic activity in fruit and seed extracts is directly related to the presence of endophytic microorganisms in these organs, where cellulase generally occurs (CARRIM et al., 2006). Therefore, the absence of this activity suggests absence of these microorganisms in *O. fieldingiana* fruits or even inadequacy of the methodology employed in this study.

Amylolytic activity was detected in CE and DCE (Starch degradation halo > 60 mm^2) of *O. fieldingiana* fruits. Two hypotheses could explain the starch degradation ability. The first one would be the expression of endogenous amylases that are related to the nutrients mobilization process on seed germination (MAR et al., 2003). The second one is the presence of endophytic microorganisms in *O. fieldingiana* fruits which are able to degrade starch (CARRIM et al., 2006). Thus, this species may be a new alternative in the search for active compounds of biotechnological and/or industrial interest.

Tab. 3 shows negative heparinase activity for *O. fieldingiana* fruits extract. However, the same extract caused 100% hemolysis in rat and mouse erythrocytes. As previously reported, species belonging to *Ouratea* genus are known to be rich in phenolic compounds (DANIEL et al., 2005; FERREIRA et al., 2006). Such compounds are widely known to promote hemolysis through hemoglobin oxidation (BUKOWSKA and KOWALSKA, 2004). In addition, the extract showed potent anticoagulant activity, which persisted for more than 24 h at 4 °C. DI STASI et al. (2002) reported anticoagulant activity for seeds of the leguminous *Caesalpinia ferrea*. A detailed study on the substance (s) which confer(s) the ability of preventing blood coagulation would reveal potential compound(s) to be used in blood coagulation studies.

The *O. fieldingiana* fruits are potentially nutritious, showing protein content in the same magnitude as legume seed crops, high dietary fiber levels, sometimes higher than conventional fiber sources in diet, as well as a low content of toxic and or antinutritional compounds. However, amino acid composition analysis, *in vivo* and *in vitro* digestibility and biological trials with rats should be performed to ensure *O. fieldingiana* safety use as food source. Several biological activities were detected in the fruits aqueous extract, such as hemolytic activity against rabbit erythrocytes, amylolytic, anticoagulant and antibacterial activity against *C. violaceum*, possibly caused by a protein. Such activities are indicative of the presence of bioactive compounds presenting pharmacological and/or industrial interest. Further studies are needed to identify and purify these compounds.

Acknowledgements

Many thanks to the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for the funding received for the research project "Bioprospecção da Caatinga Cearense com Potencial Nutricional e Bioativo" (Process 505.773/2004-0). We are also thankful to Ms Francisca Simões Cavalcanti from Laboratório de Produtos Naturais of Universidade Federal do Ceará (UFC) as well as to Mr Edson de Paula Nunes from Herbário Prisco Bezerra (UFC) for their field expeditions and botanical identification support.

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