Available online at www.sciencedirect.com
ScienceDirect
Journal of Ethnopharmacology 116 (2008) 495–500
Journal of ETHNO-PHARMACOLOGY
www.elsevier.com/locate/jethpharm

Acute and subacute toxicity of the *Carapa guianensis* Aublet (Meliaceae) seed oil

J.H. Costa-Silva [a], C.R. Lima [a], E.J.R. Silva [a], A.V. Araújo [b], M.C.C.A. Fraga [b], A. Ribeiro e Ribeiro [c], A.C. Arruda [c], S.S.L. Lafayette [a,b], A.G. Wanderley [a,b,*]

[a] *Department of Pharmaceutical Sciences, Federal University of Pernambuco, Recife, Pernambuco 50740-521, Brazil*
[b] *Department of Physiology and Pharmacology, Federal University of Pernambuco, Recife, Pernambuco 50760-901, Brazil*
[c] *Department of Chemistry, Federal University of Pará, Belém, Pará 66075-110, Brazil*

Received 19 June 2007; received in revised form 26 December 2007; accepted 27 December 2007
Available online 6 January 2008

Abstract

Carapa guianensis (Meliaceae), known as Andiroba in Brazil, has been used by Amazon Rainforest indigenous communities for treatment of coughs, convulsions, skin diseases, arthritis, rheumatism, ear infections, to heal wounds and bruises and as an insect repellent. *Carapa guianensis* seed oil (SO) was evaluated for its acute and subacute toxicity (30 days) by the oral route in Wistar rats. In the acute toxicity test, SO (0.625–5.0 g/kg, $n = 5$/sex) did not produce any hazardous symptoms or deaths. The subacute treatment with SO (0.375, 0.75 and 1.5 g/kg, $n = 10$/group) failed to change body weight gain, food and water consumption. Hematological analysis showed no significant differences in any of the parameters examined. However, in the biochemical parameters, there was an increase in the alanine aminotransferase (ALT) serum level (29%) in the group SO 1.5 g/kg. In addition, absolute and relative liver weights were increased at the doses of 0.75 g/kg (23.4 and 19.1%) and 1.5 g/kg (18.7 and 33.1%). In conclusion, acute and subacute administration of *Carapa guianensis* seed oil did not produce toxic effects in male Wistar rats. However, the increase in the ALT serum level and in both absolute and relative liver weights may indicate a possible hepatic toxicity.

Keywords: *Carapa guianensis*; Meliaceae; Acute and subacute toxicology

1. Introduction

Carapa guianensis Aublet (Meliaceae) is a well-known tree widely used as a medicinal plant in Amazon Rainforest region. In Brazil, this tree is popularly known as Andiroba, Carapá and Carapinha (Corrêa, 1984). It valued for both its timber and ethnomedicinal properties and it is recommended by United Nations Development Programme as of great pharmaceutical potential. All parts of the tree, including its seed oil are used for medicinal purposes. Traditional Amazon Rainforest communities make a medicinal soap using *Carapa guianensis* seed oil for treatment of skin diseases, arthritis, rheumatism, ear infections, to heal wounds and bruises and as an insect repellent (Hammer and Johns, 1993). They also drink the seed oil to treat coughs and convulsions (Duke and Vasquez, 1994; Branch and Silva, 1983). *Carapa guianensis* seed oil mixed with "cabacinha" fruit (*Luffa operculata* L., Curcubitaceae: 1/4 of cabacinha in 250 ml of hot oil for several hours) is rubbed into the skin to relieve arthritis and rheumatism. A spoonful of this warm maceration is gargled once a day for sore throat and ingested for cough treatment. Also, a teaspoon of the oil mixed with some honey is drunk for throat inflammation (Hammer and Johns, 1993; Duke and Vasquez, 1994). Based on the traditional use, generally 2 ml of the seed oil mixed in a small glass of warm water is taken twice or three times daily (http://rain-tree.com/andiroba.htm). Several seed oil products in many dosage forms such as cream, compound syrup, oily capsules and, more recently gel are recommended for many respiratory infections including pharyngitis, laryngitis, cough, flu, pneumonia and bronchitis as well as muscle and join injuries and skin diseases (Orellana et al., 2004).

Several studies demonstrated the antiinflammatory, analgesic and antiallergic activities of *Carapa guianensis* seed oil (Penido

* Corresponding author at: Departamento de Fisiologia e Farmacologia, CCB, Universidade Federal de Pernambuco, Av. Prof. Moraes Rego s/n, CEP 50670-901, Cidade Universitária, Recife, PE, Brazil. Tel.: +55 81 2126 8530; fax: +55 81 2126 8976.
E-mail address: almirgw@globo.com (A.G. Wanderley).

0378-8741/$ – see front matter © 2008 Elsevier Ireland Ltd. All rights reserved.
doi:10.1016/j.jep.2007.12.016

et al., 2005a,b, 2006). Other properties, such as repellent (Miot et al., 2004) and larvicide activities (Mendonça et al., 2005) were also reported.

The chemical characterization of *Carapa guianensis* seed oil revealed the presence of miristic, palmitic, oleic, linoleic (Pinto, 1956), stearic and arachidic fatty acids (Pereira et al., 1997). Some tetranortriterpenoids have been isolated from *Carapa guianensis* seeds, namely, 6-alfa-acetoxy-epoxy-azadiradione, 7-deacetoxy-7-oxogedunin, gedunin, andirobin, methyl angolensate (Pereira et al., 1999), 1,3-di-benzene carbon amine-2-octadecylic acid-glyceride, hexacosanoic acid-2,3-dihydroxy-glyceride, ursolic acid, naringenin, scopoletin, 3,4-dihydroxymethylbenzoate, 2,6-dihydroxymethylbenzoate, tetratriacontanoic acid and triacontanoic acid (Qi et al., 2004).

Notwithstanding the widespread use of *Carapa guianensis* seed oil in traditional medicine there are insufficient data about its toxicity. In the last years, our research group has investigated the *Carapa guianensis* seed oil toxic effect in order to ensure its safe use. Indeed, two recent studies performed in female rats treated *per os* with *Carapa guianensis* seed oil showed that it did not interfere on the fertility (Costa-Silva et al., 2006) and did not induce any toxic effect on pregnancy (Costa-Silva et al., 2007). Therefore, the aim of the present study was to assess the toxicity of the *Carapa guianensis* seed oil in Wistar rats. For this toxicological evaluation, some protocols are indispensable, such as acute and subacute toxicity, including hematological and biochemical analyses.

2. Materials and methods

2.1. Plant material

Carapa guianensis Aublet (Meliaceae) seeds were collected from the Amazon Rainforest (Pará, Brazil). A voucher specimen (MG170789 MA 0404), authenticated by Dra. Ely Simone Cajueiro Gurgel (Museu Paraense Emílio Goedel, Pará, Brazil), was deposited in the João Mussa Pires herbarium, at the Federal University of Pará.

2.2. Extraction

N-Hexane was added to 2 kg of previously dried and triturated *Carapa guianensis* seeds. The system was allowed to rest for 60 min and was then vacuum-filtered. The solvent was replenished until the botanical sample of oil was exhausted and then the solvent was removed under reduced pressure. The yield of extract was 34% (v/w). The apparent density of the oil was calculated (0.833 g/mL) and this was used to define the exact volume that each animal would receive. The oil was stored at −20 °C until further use.

2.3. Animals

Adult male and female Wistar rats (*Rattus norvegicus*), aged 3 and 2 months, weighing 250–300 g and 200–240 g, respectively, were obtained from Department of Physiology and Pharmacology of Federal University of Pernambuco (UFPE). They were maintained in standard environmental conditions (23 ± 2 °C; 12:12 h dark/light cycle) and water and chow (Labina®, Purina, Brazil) were available *ad libitum*. The experimental protocol was approved by Ethical Committee in Animal Experimentation of the Biological Science Center of UFPE (Process no. 23076.006909/2004-25).

2.4. Acute toxicity

Healthy rats of either sex fasted overnight, but allowed access to water *ad libitum* were randomly divided into five groups ($n = 5$/sex). The first group (control group) received water. Groups 2–5 were orally treated with *Carapa guianensis* seed oil at the doses of 0.625, 1.25, 2.5 and 5.0 g/kg, respectively. Animals were observed for general behavioral and body weight changes, hazardous symptoms and mortality for a period of 14 days after treatment. The lethal dose (LD_{50}) was estimated according to the method described by Litchfield and Wilcoxon (1949).

2.5. Subacute toxicity

The method was performed according to the OECD test guidelines with slightly modifications (OECD, 1981). Male rats were randomly divided into four groups ($n = 10$/group). Animals received water vehicle orally (control group) or *Carapa guianensis* seed oil at the doses of 0.375, 0.75 or 1.5 g/kg/day for 30 consecutive days. The body weight was recorded weekly and their food and water intake monitored daily. Animals were observed for signs of abnormalities during the treatment period. At the end of the treatment, animals were fasted overnight, but allowed access to water *ad libitum*. Then anesthetized with ether, and the blood samples were obtained by retro-orbital puncture (Waynforth, 1980), using capillary tubes for hematological and biochemical studies, with and without anticoagulant, respectively.

2.6. Hematological and biochemical analysis

Hematological analysis was performed using an automatic hematological analyzer (Coulter STKS, Beckman). The parameters included: red blood cell (RBC) count, white blood cell (WBC) count, hemoglobin (Hb), hematocrit (HCt), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and platelets count were determined (Silva et al., 2007). The differential leukocyte counting was performed with an optical microscopy after staining and, in each case, 100 cells were counted. For biochemical analysis, blood was centrifuged at $1480 \times g$ for 10 min to obtain serum, which was stored at −20 °C until determination of the following parameters: glucose, blood urea nitrogen (BUN), creatinine, aspartate aminotransferase (AST), alanine aminotransferase (ALT), total cholesterol, triglycerides, alkaline phosphatase (ALP), total and direct bilirubin, total protein and albumin. The dosages were made using Cobas Mira (Roche) automation with Boehringer Ingelheim® biochemical kits.

2.7. Morphological study

After euthanasia by excess of anesthetic, necropsy was proceeded ($n = 5$/group) for analysis of the macroscopic external evaluation of heart, liver, spleen, kidney, lungs, brain and adrenal gland. These organs were carefully removed and weighed individually. Organ weights were expressed in terms absolute and relative (g/100 g of body weight).

2.8. Statistical analysis

The results are expressed as mean ± S.E.M. Variance in data for body weights, food consumption, hematology, blood biochemistry and organ weights (both absolute and relative weights) was checked for homogeneity by Bartlett's method. If the variance was homogeneous, the data were assessed by one-way analysis of variance (ANOVA) followed by Tukey–Kramer test. When the data cannot be assumed to follow a normal distribution, non-parametric Kruskall–Wallis test followed by Dun test was performed. A probability level of less than 5% ($p < 0.05$) was considered significant.

3. Results

The results indicated that *Carapa guianensis* seed oil acute treatment by oral route at doses up to 5.0 g/kg did not produce any sign of toxicity or death in rats during 14 days of observation. Therefore, the LD_{50} could not be estimated, and it is possible more than 5.0 g/kg.

No toxicity signs or deaths were recorded during the 30 consecutive days of treatment by oral route with *Carapa guianensis* seed oil at doses of 0.375, 0.75 or 1.5 g/kg. Neither absolute body weight nor body weight gain was affected by *Carapa guianensis* seed oil administration at all doses throughout the study (Fig. 1 and Table 1). A similar lack of toxic effect was observed in the case of food and water consumption (Fig. 2).

The hematological profile of control and treated groups are presented in Table 2. There were no statistically significant differences in all of the hematological parameters analyzed.

The *Carapa guianensis* seed oil administration did not induce any changes in glucose, BUN, creatinine, AST, total cholesterol, triglycerides, ALP, total and direct bilirubin, total protein and albumin serum levels. However, an increase of 29.3% in ALT serum levels was observed within the animals treated with the highest dose of *Carapa guianensis* seed oil in comparison to control group, as summarized in Table 3.

The absolute and relative tissue weights and external macroscopic morphology were not altered by *Carapa guianensis* seed oil administration. However, a significantly increase in both

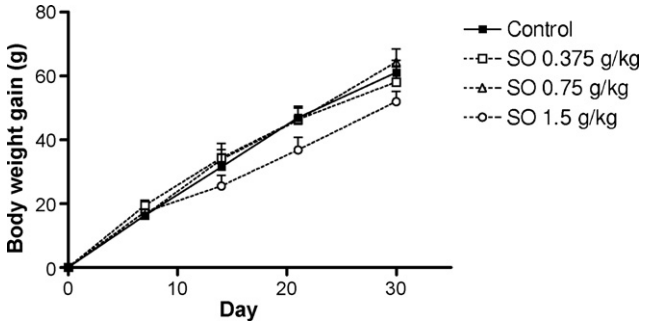

Fig. 1. Body weight gain curves of rats treated orally with *Carapa guianensis* seed oil (SO, 0.375–1.5 g/kg) by oral route for 30 consecutive days. The values are expressed as mean ± S.E.M. ($n = 10$ animals/group).

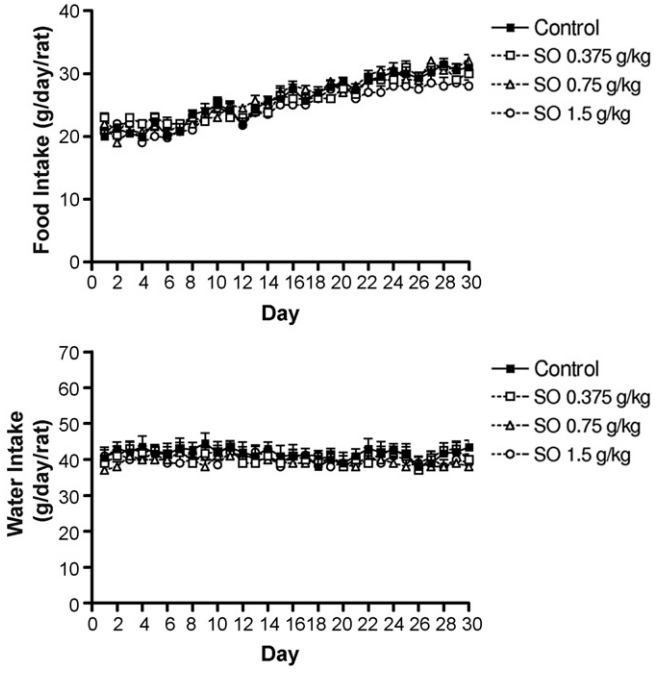

Fig. 2. Daily food and water intake of Wistar rats treated with *Carapa guianensis* seed oil (SO, 0.375–1.5 g/kg) by oral route for 30 consecutive days. The values are expressed as mean ± S.E.M. ($n = 10$ animals/group).

Table 1
Body weight (g) for male Wistar rats after 30 consecutive days of treatment with *Carapa guianensis* seed oil (SO, 0.375–1.5 g/kg) by oral route

Day	Control		SO 0.375 g/kg		SO 0.75 g/kg		SO 1.5 g/kg	
	Body weight (g)	Increase (%)	Body weight (g)	Increase (%)	Body weight (g)	Increase (%)	Body weight (g)	Increase (%)
0	304.4 ± 7.6	0	272.6 ± 7.3	0	315.3 ± 7.1	0	272.6 ± 10.8	0
7	320.7 ± 7.9	5.4	292.0 ± 8.1	7.1	331.6 ± 8.1	5.2	289.9 ± 11.8	6.3
14	335.9 ± 9.0	10.3	306.9 ± 8.5	12.6	349.1 ± 8.6	10.7	298.1 ± 11.7	9.3
21	351.3 ± 8.6	15.4	318.9 ± 7.2	17.0	361.4 ± 10.0	14.6	309.4 ± 12.2	13.5
30	365.4 ± 10.3	20.0	330.6 ± 9.2	21.3	379.6 ± 12.1	20.4	324.5 ± 11.4	19.0

The values are expressed as mean ± S.E.M. ($n = 10$ animals/group).

Table 2
Effect of the *Carapa guianensis* seed oil (SO, 0.375–1.5 g/kg) by oral route on hematological parameters in male Wistar rats treated for 30 consecutive days

Parameters	Control	SO 0.375 g/kg	SO 0.75 g/kg	SO 1.5 g/kg
RBC ($\times 10^6$ (μL)$^{-1}$)	8.0 ± 0.1	8.5 ± 0.2	8.2 ± 0.6	8.2 ± 0.1
Hemoglobin (g/dL)	14.8 ± 0.2	15.2 ± 0.2	14.6 ± 0.9	15.3 ± 0.2
Hematocrit (%)	42.7 ± 0.6	44.7 ± 0.8	42.7 ± 1.4	44.5 ± 0.4
MCV (fL)	52.7 ± 0.4	52.1 ± 0.3	53.8 ± 3.2	53.8 ± 0.7
MCH (pg)	18.6 ± 0.8	17.9 ± 0.3	18.0 ± 0.7	18.6 ± 0.2
MCHC (g/dL)	34.4 ± 0.2	33.6 ± 0.5	30.0 ± 3.2	34.0 ± 0.4
Platelets ($\times 10^3$ (μL)$^{-1}$)	1026 ± 16.7	1173 ± 47.3	1092 ± 67.2	1151 ± 49.3
WBC ($\times 10^3$ (μL)$^{-1}$)	10.9 ± 1.0	12.2 ± 1.0	11.1 ± 1.0	11.8 ± 0.9
Neutrophils (%)	23.2 ± 3.6	17.9 ± 2.8	26.8 ± 2.9	22.3 ± 2.3
Eosinophils (%)	1.7 ± 0.4	1.7 ± 0.4	1.7 ± 0.3	1.4 ± 0.2
Basophils (%)	0	0	0	0
Lymphocytes (%)	68.9 ± 3.3	74.3 ± 2.8	64.4 ± 3.5	71.7 ± 1.6
Monocytes (%)	6.2 ± 0.8	6.0 ± 0.5	7.0 ± 0.9	6.1 ± 0.9

The values are expressed as mean ± S.E.M. ($n = 10$ animals/group). RBC: red blood cell, MCV: mean corpuscular volume, MCH: mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration and WBC: white blood cell.

Table 3
Effect of the *Carapa guianensis* seed oil (SO, 0.375–1.5 g/kg) by oral route on serum biochemical parameters in male Wistar rats treated for 30 consecutive days

Parameters	Control	SO 0.375 g/kg	SO 0.75 g/kg	SO 1.5 g/kg
Glucose (mg/dL)	64.5 ± 2.2	74.2 ± 3.5	61.2 ± 3.1	56.2 ± 3.4
BUN (mg/dL)	43.8 ± 3.0	43.1 ± 1.0	42.4 ± 1.3	45.4 ± 1.8
Creatinine (mg/dL)	0.40 ± 0.02	0.40 ± 0.01	0.50 ± 0.06	0.40 ± 0.01
AST (IU/L)	237.0 ± 16.0	239.6 ± 23.4	237.8 ± 8.5	210.1 ± 4.3
ALT (IU/L)	48.8 ± 3.3	53.5 ± 3.0	48.1 ± 4.1	63.1 ± 3.2*
Total cholesterol (mg/dL)	54.7 ± 3.6	61.0 ± 3.8	59.6 ± 4.4	58.2 ± 3.1
Triglycerides (mg/dL)	53.2 ± 4.3	69.0 ± 8.8	56.8 ± 3.6	50.3 ± 5.5
ALP (IU/L)	140.8 ± 17.4	148.7 ± 14.8	111.3 ± 11.9	124.2 ± 12.6
Total bilirubin (mg/dL)	0.2 ± 0	0.2 ± 0	0.2 ± 0	0.2 ± 0
Direct bilirubin (mg/dL)	0.1 ± 0	0.1 ± 0	0.1 ± 0	0.1 ± 0
Total protein (g/dL)	6.7 ± 0.1	6.9 ± 0.1	6.8 ± 0.1	6.5 ± 0.1
Albumin (g/dL)	3.1 ± 0.1	3.0 ± 0.1	3.0 ± 0.1	3.0 ± 0.1

The values are expressed as mean ± S.E.M. ($n = 10$ animals/group). BUN: blood urea nitrogen, AST: aspartate aminotransferase, ALT: alanine aminotransferase and ALP: alkaline phosphatase.
* Significant difference from control group ($p < 0.05$ by ANOVA, followed by Tukey–Kramer test).

absolute and relative liver weight of 23.4 and 18.7%, and 19.1 and 33.1%, respectively was observed in groups treated with doses of 0.75 and 1.5 g/kg in comparison to control group (Table 4).

4. Discussion

The results of the acute toxicity study indicate that *Carapa guianensis* seed oil administration by oral route with the doses up 5.0 g/kg did not produce any sign of toxicity or death in rats, suggesting a LD_{50} above 5.0 g/kg by oral route. According to Kennedy et al. (1986), substances that present LD_{50} higher than 5.0 g/kg by oral route can be considered practically non-toxic. Therefore, it can be suggested that acute toxicity of the *Carapa guianensis* seed oil is practically null by oral route. Likewise, subacute treatment showed that *Carapa guianensis* seed oil at doses of 0.375, 0.75 and 1.5 g/kg/day during 30 days did not produce any deaths or clinical signs of toxicity. Besides, the body weight, water and food intake were not altered during the treatment period. The doses used in the present study represent up to 50 (acute) and 15 (subacute) times more than the used as an antiedematogenic agent (Penido et al., 2005a).

In previous studies, performed in female Wistar rats treated with *Carapa guianensis* seed oil *per os*, it was demonstrated that *Carapa guianensis* seed oil did not interfere in fertility parameters and in offspring development, however, an increase on the offspring motor activity was observed (Costa-Silva et al., 2006). In addition, it was verified that administration of *Carapa guianensis* seed oil did not bring about any toxic effect on pregnancy in rats (Costa-Silva et al., 2007), suggesting that *Carapa guianensis* seed oil presents low or absence of abortive effect. In present study, all hematological parameters remained under the reference range for the species (Harkness and Wagner, 1993). In the same way, the treatment with *Carapa guianensis* seed oil did not change the biochemical parameters analyzed, except for an increase in ALT serum levels in the group treated with *Carapa guianensis* seed oil 1.5 g/kg. Since the standard range for ALT serum levels for rats is 21–52 IU/L (Coimbra et al., 1995), our results provide evidence of hepatic overload.

The increase of serum transaminase enzymes (ALT and AST) levels is a good indicator of hepatocyte damage (Latha et al., 1998). The AST is also found in a large number of tissues, such as heart, lung, skeletal muscle and kidney, whereas ALT is pri-

Table 4
Effect of the *Carapa guianensis* seed oil (SO, 0.375–1.5 g/kg) by oral route on absolute and relative (g/100 g of animal) organ weight in male Wistar rats treated for 30 consecutive days

Parameters	Control	SO 0.375 g/kg	SO 0.75 g/kg	SO 1.5 g/kg
Heart				
Absolute live weight (g)	1.06 ± 0.04	0.99 ± 0.04	1.15 ± 0.02	1.01 ± 0.07
Body weight ratio (%)	0.29 ± 0.01	0.30 ± 0.01	0.31 ± 0.01	0.32 ± 0.02
Liver				
Absolute live weight (g)	10.7 ± 0.5	10.6 ± 0.5	13.2 ± 0.5*	12.7 ± 0.4*
Body weight ratio (%)	2.93 ± 0.07	3.20 ± 0.11	3.49 ± 0.11*	3.90 ± 0.12*
Spleen				
Absolute live weight (g)	0.82 ± 0.02	0.68 ± 0.03	0.73 ± 0.06	0.78 ± 0.23
Body weight ratio (%)	0.22 ± 0.01	0.21 ± 0.01	0.19 ± 0.02	0.25 ± 0.08
Kidney				
Absolute live weight (g)	1.28 ± 0.02	1.07 ± 0.06	1.29 ± 0.04	1.10 ± 0.03
Body weight ratio (%)	0.35 ± 0.01	0.32 ± 0.01	0.35 ± 0.01	0.35 ± 0.01
Brain				
Absolute live weight (g)	1.37 ± 0.05	1.28 ± 0.01	1.32 ± 0.07	1.27 ± 0.04
Body weight ratio (%)	0.37 ± 0.01	0.39 ± 0.01	0.35 ± 0.02	0.40 ± 0.01
Lungs				
Absolute live weight (g)	1.45 ± 0.06	1.86 ± 0.22	1.61 ± 0.07	1.46 ± 0.08
Body weight ratio (%)	0.40 ± 0.01	0.46 ± 0.01	0.43 ± 0.01	0.46 ± 0.02
Adrenal				
Absolute live weight (g)	0.025 ± 0.001	0.027 ± 0.002	0.029 ± 0.002	0.024 ± 0.001
Body weight ratio (%)	0.0068 ± 0.0004	0.0082 ± 0.0006	0.0076 ± 0.0008	0.0072 ± 0.0006

The values are expressed as mean ± S.E.M. ($n = 5$ animals/group).
* Significant difference from control group ($p < 0.05$ by ANOVA, followed by Tukey–Kramer test).

marily limited to liver, thus the latter is considered a highly sensitive indicator of hepatotoxicity (Al-Habori et al., 2002). The ALT in blood increases when the hepatic cellular permeability is changed or when necrosis and cellular injury occurs (Kaneko et al., 1997). Fluctuations in ALT levels are usually accompanied by an alteration of AST levels, however, AST is essentially a mitochondrial enzyme and it is not released as fast as ALT, which is cytosolic (Al-Habori et al., 2002). This could explain why an alteration was found only in ALT levels. As far as we know, increases in ALT or AST serum levels have not been reported in animals treated with extracts from other species of the family Meliaceae.

The increase in both absolute and relative liver weights of the animals treated with *Carapa guianensis* seed oil 0.75 and 1.5 g/kg observed in the present study could be associated with hepatic edema, since the edema can induce an increase on organ weight and a slight increase on the serum level of the hepatic enzymes. Further studies will be carried out to investigate the possible mechanisms involved on such effect.

Some species of the same family, including *Aphanamixis polystachya* (Gole and Dasgupta, 2002), *Trichilia roka* (Germano et al., 2001) and *Azadirachta indica* (Bhanwra et al., 2000) have demonstrated hepatoprotective activity. However, Kusamran et al. (1998) showed in male rats that dietary *Azadirachta indica* flowers (12.5%) resulted in a decrease of terminal body weight but increased the relative liver weight. In the present study there was not a body weight significant alteration but both absolute and relative liver weights were increased by *Carapa guianensis* seed oil treatment, suggesting hepatic hypertrophy. Although several substances have been isolated from *Carapa guianensis* seed oil we ignored studies that show hepatotoxicity of its components.

In conclusion, the acute and subacute oral administration of *Carapa guianensis* seed oil did not induce significant alterations in almost all biochemical, hematological and morphological parameters in Wistar rats. However, the increase in ALT serum levels and in the absolute and relative liver weights may indicate a possible hepatic toxicity. Further studies are in progress to better evaluate this finding, including hepatic histological evaluation, microsomal enzymes induction analysis and chronic treatment effects.

Acknowledgements

The authors thank Rejane de Souza Silva for excellent technical assistance and CAPES and CNPq for financial support.

References

Al-Habori, M., Al-Aghbari, A., Al-Mamary, M., Baker, M., 2002. Toxicological evaluation of *Catha edulis* leaves: a long term feeding experiment in animals. Journal of Ethnopharmacology 83, 209–217.

Bhanwra, S., Singh, J., Khosla, P., 2000. Effect of *Azadirachta indica* (Neem) leaf aqueous extract on paracetamol-induced liver damage in rats. Indian Journal of Physiology and Pharmacology 44, 64–69.

Branch, L.C., Silva, M.F., 1983. Folk medicine of Alter do Chão, Pará, Brazil. Acta Amazônica 13, 737–797.

Coimbra, I.K.S., Kosemjakin, D.A., Diniz, J.M.F., Círio, S.M., Leite, L.C., 1995. Perfil bioquímico sérico de provas funcionais hepática e renal após intoxicação experimental com extratos aquosos de folhas de *Melia azedarach* L. em ratos albinos (*Rattus norvegicus*). Brazilian Journal of Toxicology 8, 258.

Corrêa, M.P., 1984. Dicionário das plantas úteis do Brasil e das exóticas cultivadas por Manuel Pio Corrêa, vol. 1. Imprensa Nacional, Rio de Janeiro, Brazil, pp. 113–114.

Costa-Silva, J.H., Lyra, M.M.A., Lima, C.R., Arruda, V.M., Araújo, A.V., Ribeiro e Ribeiro, A., Arruda, A.C., Fraga, M.C.C.A., Lafayette, S.S.L., Wanderley, A.G., 2006. Toxicological reproductive study of *Carapa guianensis* Aublet (andiroba) in female Wistar rats. Acta Farmaceutica Bonaerense 25, 425–428.

Costa-Silva, J.H., Lyra, M.M.A., Lima, C.R., Arruda, V.M., Araújo, A.V., Ribeiro e Ribeiro, A., Arruda, A.C., Fraga, M.C.C.A., Lafayette, S.S.L., Wanderley, A.G., 2007. A toxicological evaluation of the effect of *Carapa guianensis* Aublet on pregnancy in Wistar rats. Journal of Ethnopharmacology 112, 122–126.

Duke, J.A., Vasquez, R., 1994. Amazonian Ethnobotanical Dictionary. CRC Press, Boca Raton, Florida.

Germano, M.P., D'Angelo, V., Sanogo, R., Morabito, A., Pergolizzi, S., De Pasquale, R., 2001. Hepatoprotective activity of *Trichilia roka* on carbon tetrachloride-induced liver damage in rats. Journal of Pharmacy and Pharmacology 53, 1569–1574.

Gole, M.K., Dasgupta, S., 2002. Role of plant metabolites in toxic liver injury. Asia Pacific Journal of Clinical Nutrition 11, 48–50.

Hammer, M.L.A., Johns, E.A., 1993. Tapping an Amazon plethora: four medicinal plants of Marajó island, Pará (Brazil). Journal of Ethnopharmacology 40, 53–75.

Harkness, S.E., Wagner, J.E., 1993. Biologia e clínica de coelhos roedores. Livraria Roca, São Paulo, Brazil, pp. 48–55.

Kaneko, J.J., Harvey, J.W., Bruss, M.L., 1997. Clinical Biochemistry of Domestic Animals. Academic Press, San Diego, USA, p. 932.

Kennedy, G.L., Ferenz, R.L., Burgess, B.A., 1986. Estimation of acute oral toxicity in rats by determination of the approximate lethal dose rather than the LD_{50}. Journal of Applied Toxicology 6, 145–148.

Kusamran, W.R., Ratanavila, A., Tepsuwan, A., 1998. Effects of neem flowers, Thai and Chinese bitter gourd fruits and sweet basil leaves on hepatic monooxygenases and glutathione S-transferase activities, and in vitro metabolic activation of chemical carcinogens in rats. Food and Chemical Toxicology 36, 475–484.

Latha, R.M., Geentha, T., Varalakshmi, P., 1998. Effect of *Vernonia cinerea* Less. flower extract in adjuvant-induced arthritis. General Pharmacology 31, 601–606.

Litchfield, J.T., Wilcoxon, F., 1949. A simplified method of evaluating dose-effect experiments. Journal of Pharmacology and Experimental Therapeutics 96, 99–103.

Mendonça, F.A.C., Silva, K.F.S., Santos, K.K., Ribeiro Júnior, K.A.L., Sant'Ana, A.E.G., 2005. Activities of some Brazilian plants against larvae of the mosquito *Aedes aegypti*. Fitoterapia 76, 629–636.

Miot, H.A., Batistella, R.F., Batista, K.A., Volpato, D.E.C., Augusto, L.S.T., Madeira, N.G., Haddad Jr., V., Miot, L.D.B., 2004. Comparative study of the topical effectiveness of the andiroba oil (*Carapa guianensis*) and Deet 50% as repellent for *Aedes* sp. Revista do Instituto de Medicina Tropical de São Paulo 46, 253–256.

Orellana, B.J.P., Kobayashi, E.S., Lourenço, G.M., 2004. Terapia alternativa através do uso da andiroba. Lato & Sensu 5, 136–141.

Organization Economic for Cooperation and Development (OECD), 1981. Guidelines for testing of chemicals., health effects # 407, Paris.

Penido, C., Costa, K.A., Pennaforte, R.J., Costa, M.F.S., Pereira, J.F.G., Siani, A.C., Henriques, M.G.M.O., 2005a. Anti-allergic effects of natural tetranortriterpenoids isolated from *Carapa guianensis* Aublet on allergen-induced vascular permeability and hyperalgesia. Inflammation Research 54, 295–303.

Penido, C., Costa, K.A., Costa, M.F.S., Pereira, J.F.G., Siani, A.C., Henriques, M.G.M.O., 2005b. Inhibition of allergen-induced eosinophil recruitment by natural tetranortriterpenoids is mediated by the suppression of IL-5, CCL11/eotaxin and NFkappaB activation. International Immunopharmacology 6, 109–121.

Penido, C., Conte, F.P., Chagas, M.S.S., Rodrigues, C.A.B., Pereira, J.F.G., Henriques, M.G.M.O., 2006. Antiinflammatory effects of natural tetranortriterpenoids isolated from *Carapa guianensis* Aublet on zymosan-induced arthritis in mice. Inflammation Research 55, 457–464.

Pereira, J.F.G., Teixeira, D., Gilbert, B., 1997. Chemical study of Andiroba (*Carapa guianensis* Aublet). Bolletino di Chimica Farmaceutico 136,107.

Pereira, J.F.G., Teixeira, D., Mazzei, J.L., Gilbert, B., 1999. Characterization of the chemical constituents of *Carapa guianensis* Aublet by HPLC-DAD. Bolletino di Chimica Farmaceutico 138, 77.

Pinto, G.P., 1956. Contribuição ao estudo químico do óleo de Andiroba. Boletim Técnico do Instituto Agronômico do Norte 31, 195–206.

Qi, S., Wu, D., Zhang, S., Luo, X., 2004. Constituents of *Carapa guianensis* Aubl. (Meliaceae). Pharmazie 59, 488–490.

Silva, E.J.R., Gonçalves, E.S., Aguiar, F.J.S., Evencio, L.B., Lyra, M.M.A., Coelho, M.C.O.C., Fraga, M.C.C.A., Wanderley, A.G., 2007. Toxicological studies on hydroalcohol extract of *Calendula officinalis* L. Phytotherapy Research 21, 332–336.

Waynforth, B.H., 1980. Injection Techniques: Experimental and Surgical Techniques in the Rat. Academic Press, London, pp. 3–61.