Emir. J. Food Agric. 2013. 25 (5): 394-397
doi: 10.9755/ejfa.v25i5.15510
http://www.ejfa.info/

PLANT SCIENCE

Evaluation of oil-resin activity of *Copaifera* sp. on gastric emptying in *Rattus novergicus*

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Abstract

The Copaiba oil-resin (*Copaifera* sp.) is widely used in folk medicine, especially as a healing, anti-inflammatory, antiseptic, to treat ulcers and skin diseases. Increased gastric emptying was observed with oil-resin of *Copaifera langsdorfii*, highlighting those others species from the genus of *Copaifera* can act over the gastrointestinal tract. This study evaluated the activity of the *Copaifera* sp oil-resin on gastric emptying in *Rattus novergicus*. Eighteen male rats were fasted for 24h and orally received Tween-80 and Copaiba oil-resin 100 and 200 mg/kg; one hour later they all received phenol red (PR) 0.5 mg/mL. Then, the parts of gastrointestinal tract (stomach and intestine) were analyzed. The results showed that *Copaifera* sp oil-resin at 200 mg/kg increased the gastric emptying in Rattus novergicus.

Key words: *Copaifera* sp., Gastric emptying, *Rattus novergicus*

Introduction

According to Copaiba's biologic classification, this plants belongs to Leguminosae family, subfamily Caesalpinoideae, genus *Copaifera* (Brito, 2000; Cascon, 2004; Maciel, 2002; Veiga Junior, 2005; Oliveira, 2006), and there is 72 species identified (Index Kewensis, 1996). It is natural from tropical region of Latin American and Occidental Africa (Francisco, 2005). At Brazil is possible to find 16 species exclusived find there, the most are observed at the southwest, center-west and Amazonia (Francisco, 2005).

From copaiba tree is possible to obtain its oil-resin, a transparent liquid which color ranges from yellow gold to brow, depending on the specie. However, *Copaifera langsdorfii* oil-reins are red-colored (Francisco, 2005).

Oil-resin from copaibeiras (*Copaifera* sp.) has na historical importance owing to its medical uses (Pieri, 2009), being widely used at folk medicine mainly anti-inflammatory (Rigamonte Azevedo, 2004; Araújo Júnior, 2005; Brito, 2005; Freire, 2006; Pacheco, 2006; Ramos, 2006; Silva, 2006), antibacterial (Veiga Junior, 2002, 2005; Drumond, 2004; Gonçalves, 2005; Freire, 2006; Pieri, 2007), antitumor (Maciel, 2002; Veiga Junior, 2002, 2005; Rigamonte Azevedo, 2004; Araújo Júnior, 2005; Francisco, 2005; Freire, 2006; Oliveira, 2006; Pacheco, 2006; Silva, 2006;) and healing (Brito, 2000; Maciel, 2002; Veiga Junior, 2002, 2005; Araújo Júnior, 2005; Brito, 2005; Francisco, 2005; Ramos, 2006; Silva, 2006). Gastroprotection was observed with *Copaifera langsdorfii* (0.63 mL/kg) oil-resin, besides laxative effects (Brito, 2000). Those facts highlight that other species of *Copaifera*'s genus can act over the gastrointestinal tract. The plant studied by our group is popular known as white copaiba. It had already showed antiulcerogenic activity and is in identification process and chemical characterization. Preliminary data showed that the major constituents are sesquiterpenes (trans-α-bergamotene, β-bisabolene e β-selinene), chemical compouds with known gastroprotective activity.

This aim of this study was to evaluate the activity of the *Copaifera* sp oil-resin on gastric emptying in *Rattus novergicus*.

Received 22 June 2012; Revised 03 October 2012; Accepted 18 November 2012; Published Online 01 March 2013

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Materials and Methods

The gastric emptying and small intestinal transit were assessed by the phenol red content assay, modified from the method described by Izbeki et al. (2001). Briefly, three groups of 6 male rats (242-292 g), were fasted for 24 h and orally received Tween-80 1% in water (5 mL/kg, vehicle control-VC) and copaiba oil-resin (100 and 200 mg/kg – experimental groups, Cop100 and Cop 200, respectively. One hour later, they all orally received phenol red 0.5 mg/mL in glucose 5 g% (1.5 mL/animal). After 20 min, the animals were euthanized with an overdose of sodium thiopental (100 mg/kg, i.p.) and the stomach and small intestine were removed. The small intestine was divided in the proximal (40%), medial (30%) and distal (30%) portions and each segment was homogenized in 100 mL of 0.1 N NaOH. Tissue proteins (in 5 mL homogenate) were precipitated with 0.5 mL of 20 g% trichloroacetic acid and centrifuged out (20 min, 3000 rpm). From the supernatant, an aliquot of 3 mL was added to 4 mL of 0.5 N NaOH and the concentration of phenol red was determined by absorbance at 560 nm (Biospectro SP-220 UV-VIS spectrophotometer, EQUIPAR Ltda., Curitiba, Brazil). The content of the dye in each segment was calculated and the retention of the marker was expressed as the percentage of the total amount of phenol red recovered in the four segments.

Statistical analysis

The data were analyzes by ANOVA and Tukey pos-test.

This study was approved by the Ethics Committee for Animal Research at the Federal University of Piauí (085/2010).

Results and Discussion

The rats from Cop200 group showed lower retention of phenol red ($p<0,05$) at stomach(G), proximal (P) and distal (D) portions of small intestine and higher($p<0,01$) at medial (M) portion of small intestine (G: 13,6 ± 2,8; P: 7,5 ± 1,2; M: 64,2 ± 10,0; D: 5,8 ± 0,8) (see Figure 1; Figure 2; Figure 3; Figure 4), when compared with the animals from VC group (G: 30,3 ± 3,1; P: 19,3 ± 2,1; M: 35,1 ± 3,5; D: 13,3 ± 1,9) (see Figure 1; Figure 2; Figure 3; Figure 4). The animals from Cop100 group did not presented significantly retention of phenol red (G: 37,7 ± 3,8; P: 25,5 ± 2,4; M: 26,0 ± 2,8; D: 11,3 ± 1,1) compared with VC group (see Figure 1; Figure 2; Figure 3; Figure 4).

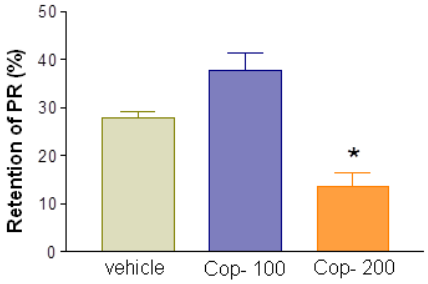

Figure 1. Shows the perceptual of phenol red at the stomach of the three groups evaluated.
*p<0,05 compared to vehicle (ANOVA and Tukey).

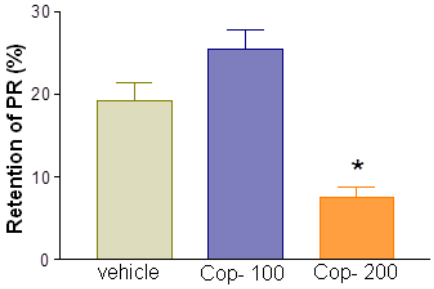

Figure 2. Shows the perceptual of phenol red at the proximal intestine of the three groups evaluated.
*p<0,05 compared to vehicle (ANOVA and Tukey post-test).

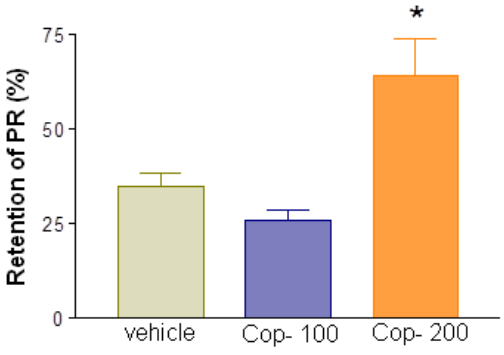

Figure 3. Shows the perceptual of phenol red at the medial intestine of the three groups evaluated.
*p<0,05 compared to vehicle (ANOVA and Tukey post-test).

The effect of a substance depends on the amount administered, but if the dose selected is below the subliminal dosing, the effect will be absent (Brunton, 2006). Depending on the nature of the effect to be measured, ascending doses may cause the effect to increase in intensity. That is why every drug has a graded dose response curve that

expresses a certain response to increasing doses of the chemical agent. (Lüllmann, 2000; Silva, 2002; Brunton, 2006; Gary, 2006; Katzung, 2006).

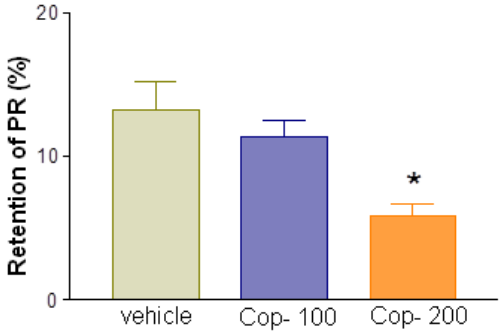

Figure 4. Shows the perceptual of phenol red at the distal intestine of the three groups evaluated.
*p<0,05 compared to vehicle (ANOVA and Tukey post-test).

In fact, the intensity of a pharmacologic response is proportional to the number of receptors with which a drug interacts (Silva, 2002; Katzung, 2006). The receptors denote the component of the organism with which the medicament is presumed to interact, modifying the function of the body component and thereby initiating biochemical and physiological changes that are characteristic of the response to the drug (Lüllmann, 2000; Brunton, 2006; Gary, 2006; Katzung, 2006). So, even if at a certain dose the drug produces no response, response can be achieved with higher doses (Lüllmann, 2000; Silva, 2002; Brunton, 2006; Gary, 2006; Katzung, 2006).

The elimination of the copaiba of the human body is made by the lungs, kidneys, sebaceous and sweat glands (Veiga Junior, 2005; Opção Fênix, 2011). In large quantities it can cause side effects like gastrointestinal irritation, vomiting, nausea, salivation, diarrhea, irritative effect in the peritoneum, causing adhesions and abscess formation and cavitary central nervous system depression. In normal doses it has anti-inflammatory effects without causing gastric damage. Thus, it becomes an agent clinically safe and potentially useful (Veiga Junior, 2007; Pinto, 2002; Opção Fênix, 2011).

As a drug, the oil-resin of Copaifera sp. is not different. Scientific studies showed that administration of 0.4 ml of the oil-resin via transdiaphragmatic caused diarrhea (Sousa, 2000). Researches with commercial copaiba oil in rats showed the occurrence of diarrhea, weight loss and irritating action on the behavior of rats at doses of 0.63 ml / kg. It is also known that gastrointestinal irritation, diarrhea, drooling and central nervous system depression are adverse effects of high doses of the oil (Veiga Junior, 2005).

Therefore, due to the proven link between the plasma concentration of the drug (which depends directly on the dose) and the proportion of the desired therapeutic effect (Brunton, 2006), the oil-resin of *Copaifera* sp., at 200 mg/kg, increased significantly the speed of gastric emptying in *Rattus novergicus*, however, at 100 mg/kg no kinetic activity was showed by *Copaifera* sp. oil-resin.

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