

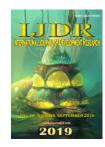
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PHARMACOLOGICAL ACTIVITY OF THE FLAVONOID PECTOLINARIN FROM THE LEAVES OF LANTANA CAMARA (VERBENACEAE)

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ABSTRACT

The leaves of *Lantana camara* were submitted to preliminary phytochemical study obtaining essential oil and methanolic extract. In the analysis of the essential oil (hydrodistillation), they identified cariophilene (7.3%), Germacrene B (37.5%) and Germacrene D (21.7%) as the most relevant of the nine components obtained. The ethyl acetate (EtOAc) revealed from the partition of the ethanolic extract (EtOH) of the Leaves of L. camara was subjected to filtration and column chromatography (CC) resulted in lavone, unheard of in the species. The structure was defined as 1 H and 13 C (1 D and 2 D) by nuclear magnetic resonance (NMR) and mass spectrometry, similar to 5'-O- α -L-rhamminopyranosyl-1'- β -D- glucopyranosyl-4', 6-dimethoxyflavone, pectolinarin. This compound has been subjected to biological tests, larvicide, acetyl cholinesterase, antioxidant and cytotoxicity. The compound isolated at 600 ppm, caused low mortality. A complete analysis of 95% of the deaths during 48 hours of exposure of the worms to the extract showed effective larvicidal action. The antioxidant activity of the compound, in the face of the free radical sequestration method DPPH, presented an LD50 0.29 mg. mL-1. Regarding its toxicity, it showed a lethal concentration (LC50) of 35.7 ppm, indicating high toxicity, and remaining active for more than 48 hours.

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INTRODUCTION

The plant species *Lantana camara* (Verbenaceae) was initially studied by Linnaeus in 1753, which described seven species, six of them originating in South America and one from Ethiopia. It is estimated that the current approximate number of species is 150, distributed in 50 countries (GHISALBERTI, 2000). Occupying tropical regions, sub, this vegetal species is a plant that develops in abandoned areas in both dry and humid areas and often grows in valleys and slopes, competing with grasses for ecological dominance (SHARMA; SHARMA, 1989). In Brazil, it is known by several other popular names, amongthem: "chumbinho", "fennel", "cambará-de-Espinho", "cambará-de-cheiro", "cambará-de-lead", "cambará-yuba", "cambará-de-cheiro", "cambará-de-lead", "cambará-vermelho",

"cambará-kid", "cambará-real", "large-leaf cambará". "lantana", "lantana-espinhosa" (LORENZI, $1982 \cdot$ TOKARNIA et al., 2000). It is a plant of the sun, does not require the soil, adapting easily to the environments that meet, resistant to pruning, springing quickly and flourishes practically all year (CORREA, 1984), adapting as invasive plants, expanding in unusable areas (WATANABE, 2005; BRITO et al., 2004). Several studies have been conducted and described in the literature, demonstrating the effects of extracts obtained with vegetables of the genus Lantana on various organisms, including mosquitoes and their larvae. In the literature, it is cited as a toxic plant, capable of causing photosensitization (a disease caused by toxins) in cattle or sheep, although not all species possess this property (BRITO et al., 2004; TOKARNIA et al., 1999). A plant that is often used

in folk medicine, such as antiseptic, against hemorrhages, flu, and in the treatment of diarrhea (SAGAR et al., 2005). Also known are allelopathic properties, plant-released, and watersoluble substances (GORLA; PEREZ, 1997), and currently stands out with significant repellant effects against Aedes mosquito larvae (IANNACONE; LAMAS, 2003). According to Ahmed (1984), many studies have been conducted to search for fundamental methodologies to obtain less aggressive substances to humans, and the environment has grown considerably in recent years. The search for alternative methods for chemical control against harmful insects or vectors of diseases is the use of plant extracts and natural substances, which currently reveal their potentiality in the control of adult mosquito and the larva of Aedesaegypti toxic. environmentally Among them, the nicotine (Nicotianatabacum), Rotenone (Lonchocarpus sp.) and Pyretropyrethrin (Chrysanthemum cinerariaefolium), were the first compounds obtained from vegetables and marketed as insecticide around the years 50. From this plant species, it was possible to extract, through its leaves, essential oils, which are volatile substances, extracted through distillation by steamdragging (hydro distillation). Within this perspective, the present work presents the composition of the essential oil of its leaves and biological activity and the data of ¹H and ¹³C NMR of the isolated flavonoid, unprecedented for this species, which may be of great value in the search for potential products with pharmacological properties.

MATERIALS AND METHODS

To obtain essential oil and ethanolic extracts was made a leaves selection from L. camara species in 2016 at the Vale do Alemão unit in Pernambuquinho, Guaramiranga, Ceará(S4 ° 12 ' 22.7 "and W38 $^{\circ}$ 57 ' 00.8 "). The exsicata of the biological material was sent to the Herbarium Prisco Bezerrain the botany department of Pici Campus at Ceará Federal University (UFC), later identified as EAC0060819. The leaves were cut and disposed in containers to be weighed and soon after taken to the incubator Greenhouse EIP-010 (Caltech®) with circulating air to avoid saturation with water vapor that is being detached from the material that is drying and exposed to a constant temperature of 50 °C for 48 hours for drying. Soon afterward, they were weighed again on the scale of precision model BL3200H (Marte®) shortly after the material was crushed into a blender (Philips®) and stored in a 1000 ml becker. Column adsorption chromatography's were made using 60 silica gel from Merck (Ø µm 63-200) Art. 7734. The size of the columns (length and diameter) varied according to the amount of sample to be submitted to chromatography and employed. The LC/MS chromatographic adsorbent analysiswas carried out in an Acquity UPLC (Waters) system, coupled to a quadrupole/flight time system (QtoF, Waters) belonging to the Brazilian agricultural research company-EMBRAPA. The chromatographic races were carried out in a column Waters Acquity UPLC BEH (150 x 2.1 mm, 1.7 a), fixed temperature of 40 °C, moving water phases with 0.1% of formic acid (A) and acetonitrile with 0.1% of formic acid (B), gradient varying from 2% to 95% B (15 min), flow of 0.4 mL/min and injection volume of 5 uL. The ESI-mode was acquired in the range of 110-1180 Da, source temperature fixed to 120 ° C, desolvation temperature 350 °C, flow of the gas destabilization of 500 L.h⁻¹, extraction cone of 0.5 V, the capillary voltage of 2.6 kV. The ESI + mode was acquired in the range of 110-1180 Da, fixed source temperature of 120 °C, desolvation temperature 350 °C, flow of the gas destabilization

of 500 L/h and capillary voltage of 3.2 kV. Leucine Encephaly was used as a lock mass. The acquisition mode was MSE. The Masslynx 4.1 software (Waters Corporation) controlled the instrument. The volatile composition analysis of the essential oil was performed in GC/MS Shimadzu/OP2010 using a capillary column RTX-5 (30 m x 0.25 mm, 0.25 A-micrometer thick film) and the drag gas is helium. For analysis of the temperature of the essential oil, the furnace was programmed of 40-180 °C at a rate of 4 °C.min⁻¹, after 180-280 °C at a rate of 20 °C.min⁻¹ and maintained at 280 °C for 10 min. For the analysis of the fixed temperature of the oil furnace was programmed of 80-280 °C At a rate of 5 °C.min⁻¹, then from 280-300 °C at a rate of 20 °C.min⁻¹ and kept at 300 °C for 5 min. The Kovats retention indexes were calculated using a series of standard N-alkane (C7-C30). The identification of the compounds was performed by comparing their mass spectra with those of the NIST08 library, retention indexes, and published data (ADAMS, 2007).

Nuclear Magnetic Resonance Spectroscopy (NMR)

The proton nuclear magnetic resonance spectra (¹H NMR) and carbon-13 (NMR $^{13}\tilde{C}$), uni and two-dimensional, were obtained in the Bruker spectrometer, models DRX-300 and DPX-500, operating in the frequency of 300 and 500 MHz for proton and for carbon-13, 75 and 125 MHz, respectively. The deuterated solvents used in the dissolution of the samples and obtaining the spectra were Chloroform (CDCl₃), acetone $[(CD_3)_2CO]$, methanol (CD₃OD), and pyridine (C₅D₅N). The multiplicities of the absorptions were indicated according to the Convention: s (Singlete), D (doublet), DD (double doublet), T (Triplete), DDD (double doublet doublet) hep (Heptet) and M (Multiplete). The hydrogenation pattern of the carbons in ¹³C NMR was determined by using the DEPT (Distortion less Enhancement by Polarization Transfer) technique, with a nutation angle of 135°, CH and CH₃ with an amplitude as opposed to CH₂. The non-hydrogenated carbons were characterized by the subtraction of the signals of the spectrum BB (Broad Band) and DEPT and the second Convention, the methyl, methylenic and methyl terminology, was applied to the groups CH₃, CH₂, and CH, respectively.

Preparation of the ethanolic extract of the leaves of Lantana camara

The material was separated 100 g from the sample and added in 450 ml of hydrated ethyl alcohol (ethanol) 92.8%, remaining at rest from 24 to 48 hours. The ethanolic extract was obtained after several successive extractions at four-day intervals. Subsequently, they were filtered and placed in a becker. The extractive solutions were subjected to the process of a rotative evaporator, at a temperature of 60° to 70°C, under pressure from 500 to 750 mmHg, with rotation from 40 to 80 r.p.m, where it resulted in a crude ethanolic extract of 18.4 with \pm 0.1%. The essential oil of the leaves of *L*. *camara* was extracted by the Hydrodistillation method of the adapted Cleavenger type (TISSERAND, 1995). The process consists in vaporizing the essential oil by a water vapor current, coupled to the round-bottomed flask of 2000mL, allows the separation of oil and water (hydrolat) and cooling of the oil, avoiding the decomposition of it. The 310g of leaves were placed in the round-bottomed flask with deionized water and added to the warming blanket. The extraction time of 2h from the boiling of the sample. The volatile oils have higher vapor voltage than water, leaving at the top of the distiller, and then passing through a cooling. So the water and oil are condensates. In this output product can be seen the difference of two phases, oil at the top and in the lower water; They are separated by a decanting process. During distillation continued to add water through the vertical flow condenser. Then the water was removed and then the oil. The organic phase was dried with anhydrous sodium sulfate (Na₂SO₄), and the mixture was filtered in a ruched paper, washed with CH₂Cl₂ (dichloromethane) and then taken to the rotary evaporator for the removal of the solvent. The material obtained was 1, 2g, with a density of 0, 879g. mL⁻¹, and mass yield of 0.8%.

Antioxidant activity of essential oil from *Lantana camara* by DPPH

According to Hegazi and collaborators (2003) and Fonseca (2012), this method consists in evaluating the antioxidant capacity via free radical sequestrating activity of 2.2-diphenyl-1-picrylhydrazyl (DPPH). The DPPH radical has a purple coloration absorbing at a maximum wavelength of approximately 520nm. By the action of an antioxidant (AH) or a radical (R[.]) species, the DPPH is reduced by forming diphenyl-picrylhydrazine, yellow staining, with consequent disappearance of absorption, and may be monitored by the decrease of absorbance. The measurements were made by adding to the sample a mixture containing 1ml of the sample at various concentrations in ethanol $(1\text{mg.mL}^{-1}; 0, 5\text{mg.mL}^{-1}; 0,$ 25mg.mL^{-1} ; 0, 0125 mg. mL $^{-1}$; 0, 0625 mg. mL $^{-1}$ and 0, 03125mg. mL⁻¹) and 1mL of DPPH 60 μ M. Shortly after the absorbance was measured at 520nm, for the white was made 1mL of ethanol without sample. The methodology used for the antioxidant activity was performed in triplicate for each concentration analyzed (TEPE et al., 2005).

Cholinesterase inhibition test

For the cholinesterase inhibition test, reagents per pit were used: 25 μ L of acetylthiocholine iodide (15 mM), 125 μ L of 5.5 '-dithiobis-[2-nitrobenzoic] in Tris/HCl solution with 0.1M NaCl and 0.02 M MgCl₂.6H₂O, 50 M L of the Tris/HCl with 0.1% solution bovine serum albumin, 25 μ L of the flavonoid sample dissolved in ethyl acetate. The preparation of the samples was flavoid (2mg.mL⁻¹). The pattern was physostigmine, with a dilution curve from 2 mg mL⁻¹. The inhibitory activity of the enzyme acetyl cholinesterase (AChE) was measured in plates of 96 wells of flat fungus using Elisa BIOTEK Reader; model ELX 800, software "Gen5 V 2.04.11", based on the methodology described by ELLMAN et al. (1961). Data were standardized to estimate non-linear regression curves by Graph Pad Prism v 5.01 statistical software.

Toxicity activity of the pectolinarin through Bioassay with saline (*Branchipus stagnalis*)

Artemia Salina Leach, of the order Anostraca, is a species of Microcrustacean that feeds primarily on bacteria, single-celled algae, small protozoa and debris dissolved in the aquatic environment. Being found in salted water and commonly used as food for fish. The toxicity of these micro crustaceans occurs through the high level of concentration of these animals in the digestive tract of individuals as the filtration rate decreases with the increase of particle concentration, being accumulated and interfering with the normal process of your heartbeat. Another effect of high concentrations is that they can pass

directly through the digestive tract without suffering digestion, making the individual undernourished (NASCIMENTO et al., 2008). The toxicity of the flavonoid was tested in the model of saline (Branchipus stagnalis) based on the technique described by Meyer et al. (1982) adapted. The cysts (larvae) were placed in a becker with saline water, under aeration, artificial illumination and temperature control (26-30 °C) and incubated for 24 hours for the hatching of eggs. It was used 18mg of the flavonoid from the leaves of *L.camara* dissolved in 18mL of dimethylsulfoxide (DMSO) 1%. The sample was dissolved in the ultrasonic bath to prepare the mother solution. From the mother solution, 5mL were dissolved in saline water until the concentrations of 31.2 were obtained; 62.5; 125; 250; 500 ppm. The assay was performed in triplicate samples, and saline water was used as a negative control. With the aid of a Pasteur pipette, ten larvae were transferred to each test tube. After 24 and 48 hours in contact with the suspension of the compound, the number of dead larvae was counted. Those larvae that remained motionless for more than 10 seconds after gentle agitation of the tubes were considered dead. The calculation of the mean lethal concentration (LC₅₀) of the flavonoid was made from the five concentrations studied using the PROBIT log-dose analysis program (FINNEY, 1952).

Evaluation of larvicide Activity the pectolinarin from *L.camara* against *Aedesaegypti*

The leaves were washed in current distilled water, dried in air circulation greenhouse with a temperature of 50 ° to 70 °C degrees, during 48 hours. After drying, they were weighed, crushed, and subjected to extraction by maceration in ethanol. The extract was filtered and rotated, with rotation from 40 to 80 rpm and temperature from 50 ° to 70 °C degrees. The eggs of Aedes spp. were collected in the Barreto Park, located in the neighborhood of Saint JohnofTauape, and on the trail of poop, in the neighborhood of Cocó, both in the municipality of Fortaleza-CE, with views of acquiring samples of Ae. aegypti and Ae. albopictus of geographically distinct populations. The realization of such collections was through oviposition traps (known as ovitrampas), prepared according to Fay and Eliason (1966). The positive ovitramps were sent to the laboratory of Medical Entomology of the Department of Pathology and Legal Medicine of the Federal University of Ceará, where they were immersed in distilled water to induce hatching of eggs, whose resulting larvae were Transferred to 300 mL plastic containers, fed with soy protein-based ration and kept at a temperature of 25 ± 2 °C. The resulting pupae were distributed in plastic containers of 100 mL, which were inserted inside the cages, adapted to the maintenance of the emerging winged forms. Once the adult stage was reached, mosquitoes, identified according to the specific classification key (FORATTINI, 1995), were fed a 10% sucrose solution, soaked in cotton, and for the females ' blood repasture were used Quails anesthetized (Coturnixcoturnix). Mosquitoes were exposed to a controlled photoperiod, with 14 hours of light and 10 hours of darkness. Larvae (20 specimens/sample) of the 3rd and 4th stages of offspring generation from colonies kept in the laboratory of Ae. aegyptiand Ae. albopictus were used for the larvicidal assays with the sample obtained, according to the parameters established by the World Health Organization (WHO, 1981). The samples were treated with four different concentrations (400, 600, 700, 900 ppm) of the isolated flavonoid, carried out in triplicate, from which the lethal concentrations of the product (s) responsible for mortality were estimated. 50% (LC₅₀) of the specimens tested.

RESULTS AND DISCUSSION

Through the data it was possible to calculate the antioxidant activity of the essential oil of L. camara and the isolated compound, making comparisons with vitamin C and Trolox, through an equation of the line, to obtain 50% of activity (IC_{50}) , as shown in Table 1. According to the methodology performed (HEGAZI; EL HADY, 2002), the obtained oil presented moderate antioxidant activity, in comparison with the isolated flavonoid and the positive controls Trolox and vitamin C. Based on the data, an antioxidant action of the oil with IC_{50} of 0.44mg.mL⁻¹, and of the flavonoid with 0.29 mg.mL⁻¹ was observed, when compared to the positive patterns of ascorbic acid (0.27mg.mL⁻¹) and Trolox (0.23mg.mL⁻¹).

Table 1. Percentage of free radical sequestration DPPH for 50% of activity (IC₅₀) of CC essential oil and flavonoid

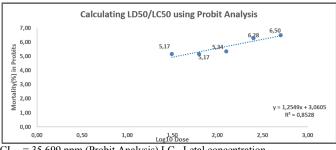
Treatment	Concentration IC ₅₀ (mg.mL ⁻¹)		
Trolox	0.23	±0.02	
Ascorbicacid	0.27	± 0.01	
Essentialoil	0.44	±0.03	
pectolinarin	0.29	±0.01	

After normalization of the data, the test of the non-linear regression curve was performed, obtaining the following results, as shown in Table 2. It was observed, in the evaluation of the enzyme acetyl cholinesterase, that the flavonoid obtained a value of IC_{50} near the standard sample. The data had an adjustment to the regression model in 97%.

Table 2. Nonlinear regression curve by GraphPad Prism v 5.01 statistical program

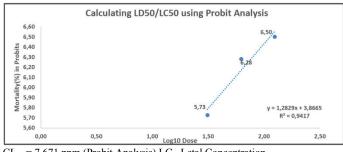
Sample	IC ₅₀	Standard error	R ²
Standard	1.15 mg.mL ⁻¹	0.05	0.99
pectolinarin	1.20 mg.mL ⁻¹	0.06	0.97

According to the results of the bioassay with saline (Branchipus stagnalis), in the concentrations tested, in the incubation period with artificial light and temperature controlled between (26 - 30 °C) for 24 hours, presented a lethal concentration (LC₅₀) of 35.699 ppm, according to the methodology adopted, it is suggested that the flavonoid, from this assay, highlights high toxicity as shown in Graph 1. In 48 hours, the compound remained with its high toxicity, LC₅₀ of 7.671 ppm, killing a high number of larvae (Graph 2), stating that the lethal concentration of the extract does not need to be elevated to eliminate large quantities of larvae (Table 3). The toxicity test with saline is a biological assay used continuously for testing toxicities because it is fast, reliable, low cost and has demonstrated good correlation with various biological activities (MEYER et al., 1982), as an activity Antitumor (MCLAUGHLIN, 1991a; MCLAUGHLIN et al., 1991b, 1993). According to the Hodge scale and Sterner extracts or compounds with $LD_{50} \le 50$ ppm are considered highly toxic substances. The experiments with repeated samples (triplicate) recur that plants of the genus Lantana have an accumulative effect and when administered in doses that correspond to a lethal dose causes severe intoxications (TOKARNIA et al., 1999). For the bioassays, larvae of the third and fourth stages of Ae.aegypti were used, exposed to four different concentrations of the extracts. The control group consisted of deionized water and diluent. Four replicates per treatment were used, with 20 larvae per repetition. The flavonoid analyzed at a concentration of 600ppm, which caused mean mortality above 90% of the larvae tested and complete analysis of 95% of death, up to 48 hours of exposure of the worms to the aforementioned compound, (Table 4). Through the study, Probit (Excel), LC₅₀ of 136.9 ppm was verified. It should be emphasized that species of the family Verbenaceae, with a bitter flavor, have insecticidal activity and pesticides, which increases the interest to be investigated for actions against the Ae. aegypti.



CL₅₀ = 35.699 ppm (Probit Analysis) LC- Letal concentration.

Graph 1. Concentration-response curve of the mortality of saline to flavonoid after 24 hours



CL₅₀ = 7.671 ppm (Probit Analysis) LC- Letal Concentration.

Graph 2. Concentration-response curve of the mortality from saline to the flavonoid after 48 hours

Table 3. The average mortality of Artemiasalina according to the concentrations of the extract

concentration(mg/mL)	Number of cysts (larvae) exposed*	Deadcysts (24h)	Deadcysts (48h)
0.5	30	28	30
0.25	30	27	30
0.125	30	19	28
0.0625	30	17	27
0.0312	30	17	23
(Negative control/saline water)	30	0	0

*Process by triplicate

Table 4. Larvicide assay with flavonoid in 48 hours

Concentration(ppm)	Numberoflarvaeexposed*	Deadlarvae (48h)*
400	60	53
600	60	54
700	60	52
900	60	54

The larvicide effect of the flavonoid studied may be associated with the various other compounds present in the species, corroborating the reports of Aguiar-Menezes (2005), where the author also draws attention to the action of the active principles present in the plants which are usually composed of a complex mixture of substances that can cause inhibition of insect feeding or hinder their growth, development, reproduction and behaviour, also present toxic actions capable of acting on the Central nervous system, which can cause the death of insects, and can still serve as an antifood agent, preventing insects from initiating feeding, thus contributing to their death. These compounds may still be acting alone or in synergism, a fact that can be proven with reports made by Bessa (2007) where the author states that the crude extract may present in some cases more significant effect when compared with their respective Fractions where some compounds are isolated, and in other situations the isolation of these compounds facilitates the knowledge of the chemical structure of the plants and contributes to the production of semisynthetic derivatives, and these can act with a better performance in relation to the potency, stability or safety of the compound. In GC/MS and FID analysis, nine compounds were identified, representing 84.1% of the total components detected from the Lantana camara leaves. The chemical composition of the oil is presented in Table 5.

Table 5. Constituents of the essential oil of lead cambará leaves by GC/MS

Compostos	RI	Porcentagem %	Identificação
α - pineno	7.56	1.2	RI, MS, Literature
β - pineno	8.94	1.0	RI, MS, Literature
γ - terpineno	11.84	3.1	RI, MS, Literature
Copaeno	23.25	5.0	RI, MS, Literature
Cariofileno	24.72	7.3	RI, MS, Literature
α – cubebeno	25.74	2.1	RI, MS, Literature
Germacreno D	26.74	37.5	RI, MS, Literature
Germacreno B	27.23	21.7	RI, MS, Literature
(+)-δ-cadinene	28.06	5.2	RI, MS, Literature

The three main compounds that compose the aroma of volatile oil were sesquiterpenes, presenting the following composition: Cariophilene (7.3%), Germacrene B (37.5%), and Germacrene D (21.7%).

Structural determination of the compound isolated

The mass spectrum of the glycosylated substance 1 revealed the presence of fragments corresponding to the molecular ions, noting in all close similarity with that of pectolinarin ([M]+ 622, $C_{29}H_{34}O_{15}$), Figures 1 and 2. In the MS of 1, the base peak of m/z 622 represents the aglicone unit with molecular formula C₂₉H₃₄O₁₅. The glycoside nature of these substances as indicated by the number of oxymethyl and oxymethyl signals observed in ¹H and ¹³C NMR spectra and confirmed by 2D Homonuclear (COSY) and Heteronuclear (¹H-¹³C-HMQC and ¹H-¹³C-HMBC) spectra), allowing the characterization of the structures of these glycosides according to the literature (WIDYOWATI et al., 2016), as shown in Table 6. The compound (1) was obtained as a pale brown powder with the molecular formula C₂₉H₃₄O₁₅ was determined by HR-ESI-MS at m/z 622.1898 [M + H] + (calc. for 623.1898). The ¹H NMR Spectrum (Table 1) displays methyl signals due to rhamnose in δ H 1.17 (d, J = 6.2 Hz), two methylenic oxygenated protons of glucose equivalents in $\delta_{\rm H}$ 3.9 (m) (br d, J = 9.9 Hz), two singletone signals for two methoxy groups in $\delta_{\rm H}$ 3.76 (s) and 3.85 (s) (3H, s), two anomeric protons in $\delta_{\rm H}$ 4.25 (s) and 4.82 (d) (d, J = 7.2 Hz), two aromatic protons in 6.88 (s) and 6.92 (s), and a coupling system of type AA ' BB ' in $\delta_{\rm H}$ 8,022 (d, 8.9) and 7.15 (d, 8.9). The Carbon resonance spectrum (¹³C NMR), (Table 6) of 1 showed 29 carbon signals, which were classified by the values of chemical displacement and quantum heteronuclear coherence (HSQC) single as two sugars (ramnosis: δ_C 17.78, 68.32, 70.44, 70.82, 72.06 and 100.45), and glucose: 66.02, 69.57, 73.20, 75.78, 76.48 and 100.45),

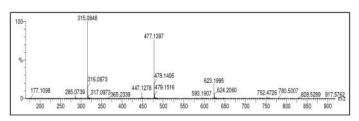


Figure 1. High resolution mass spectrometry with electrospray ionization (HRESIMS)

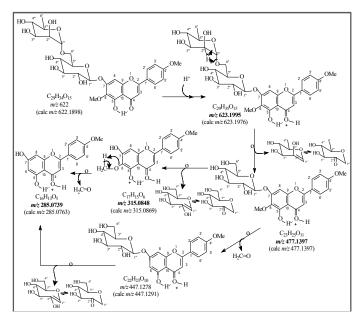


Figure 2. Fragmentation proposed for the isolated structure

Table 6. Spectroscopic data of the structure (1)

	HSQC		HMBC	
1				
	$\delta_{\rm C}$	$\delta_{\rm H}$	$^{2}J_{\mathrm{CH}}$	$^{3}J_{CH}$
С				
2 4	164.23	-	H-3	
4	182.42	-	H-3	
5	152.29	-		
6	132.81	-		H-8; MeO-6
7	156.57	-	H-8	
9	152.30	-	H-8	
10	106.01	-		H-3; H-8
1'	122.80	-		H-3; H-3'/H-5'
4'	162.51	-		H-2'/H-6'; MeO-4'
CH				
3	103.43	6.88 (s)		
8	94.46	6.92 (s)		
2',6'	128.52	8.022 (d, 8.9)		
3',5'	114.85	7.15 (d, 8.9)		
1''	100.45	4.82 (d)		
2"	73.20	3.35		
3"	75.78		Н-3''	
4''	69.57			
5''	76.48			
1'''	100.45	4.25 (s)		
2'''	70.44			
3'''	72.06			
4'''	70.82			
5'''	68.32		3H-6'''	H-1""
CH_2				
6''	66.02	3.90 (s)		
CH_3				
6'''	17.78	1.17 (d, 6,2)	H-5'''	
MeO-6	60.43	3.76 (s)		
MeO-4'	55.64	3.85 (s)		
HO-5	-	12.92 (sl)		
AcO-2""	-	-	-	-
AcO-3""	-	-	-	-

two carbon methoxylated (δ_C 55.64 and 60.43), four methyl carbons of type sp² (δ_C 103.43, 94.46, 114.85 (2xC) and 128.52 (2xC)), nine quaternary carbons of type sp² (δ_C 106.01, 122.80, 132.81, 152.29, 152.30, 156.57, 162.51 and 164.23) and a carbonyl carbon in δ_C 182.42.

Conclusion

This work reported the study of the plant species Lantana *camara*, which from the ethanolic extract of the leaves, using several chromatographic procedures, allowed to isolate a bioid of flavone, pectolinarin. It is also noteworthy that the aforementioned glycosylated flavonoid is unprecedented in this plant species. From the aerial parts of the species was extracted its volatile composition, which through the GC/MS technique, it was possible to identify the presence of three terpenoid compounds, caryophyllene (7.3%), germacrene B (37.5%), and the germacrene D (21.7%), which are probably responsible for their intense aroma. In the antioxidant test made by the free radical sequestration method (DPPH), the compound above presented an IC_{50} of 0.29 mg.mL⁻¹, very close to the definite trolox pattern. In the toxicity activities, both in Artemiasalina and as in arbovirus A. aegyptlarvae, was observed an $LD_{50} \le 50$ ppm, that according to literature is considered very high toxicity. By inhibitory activity of the acetyl cholinesterase enzyme, the compound presented an IC_{50} of 1.2mg.mL⁻¹, very close to the definite pattern used. It can be affirmed that, through the data presented in this work, this plant material has a biological potential of considerable interest, both for the chemistry of natural products and pharmacology.

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