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The *Jatropha mollissima* (Pohl) Baill: chemical and pharmacological activities of the latex and its extracts

A *Jatropha mollissima* (Pohl) Baill: caracterização química e atividades farmacológicas do látex e seus extratos

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Abstract

The *Jatropha mollissima* (Pohl) Baill is an autochthonous shrub native to Caatinga vegetation, which is found in much of the semi-arid region of the Brazilian Northeast. It is used in popular culture for therapeutic purposes. The latex of *Jatropha* and its extracts were evaluated for chemical composition, identifying the main secondary metabolites and their pharmacological activities by using qualitative and quantitative methods. Its toxicity was investigated through the acute toxic dose in Wistar rats. The antioxidant action was investigated according to the photocolorimetric method of free radical DPPH. Their antibacterial action was investigated through minimum inhibitory concentration and minimum bactericidal concentration. Phenols, tannins, flavonoids and saponins were detected in latex and other extracts, except the aqueous extract. Both latex and ethanolic extracts presented moderate antioxidant activity, as well as anti-bacterial activity against Gram-positive (*Staphylococcus aureus, Streptococcus agalactiae, S. pyogenes, S. mutans and Enterococcus faecalis)* and Gram-negative strains (*Shigella flexneri, Escherichia coli, Klebsiella pneumoniae, Salmonella typhimurium and Pseudomonas aeruginosa*) in various dilutions along with the hydroalcoholic extract. *J. mollissima* latex showed low acute toxicity in rats. Therefore, the results obtained in this research corroborate the popular use of *J. mollissima* and its therapeutic potential.

Key words: Brazilian semi-arid. Biological activities. Euphorbiaceae. Medicinal plants.

Resumo

O pinhão-bravo (*Jatropha mollissima* Pohl Baill.) é um arbusto autóctone da vegetação de caatinga, encontrada em grande parte do semiárido do Nordeste brasileiro, utilizado na cultura popular com fins terapêuticos. O látex do pinhão-bravo e os extratos dele derivados foram avaliados quanto à composição química, identificando os principais metabólitos secundários a partir de métodos qualitativos e

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quantitativo, e quanto as suas atividades farmacológicas. Foram pesquisadas toxicidade, através da dose tóxica aguda em ratos Wistar, ação antioxidante, de acordo com o método fotocolorimétrico do radical livre DPPH, e ação antibacteriana, através da concentração inibitória mínima e da concentração bactericida mínima. Foram detectados fenóis, taninos, flavonoides e saponinas no látex e demais extratos pesquisados, exceto o extrato aquoso. Tanto o látex quanto o extrato etanólico apresentaram moderada atividade antioxidante, como também evidenciaram atividade antibacteriana contra as cepas Gram-positivas (*Staphylococcus aureus*, *Streptococcus agalactiae*, *Streptococcus pyogenes*, *Streptococcus mutans* e *Enterococcus faecalis*) e Gram-negativas (*Shigella flexneri*, *Escherichia coli*, *Klebsiella pneumoniae*, *Salmonella typhimurium* e *Pseudomonas aeruginosa*), em várias diluições, juntamente com o extrato hidroalcoólico. O látex do pinhão apresentou ainda baixa toxicidade aguda em ratos. Logo, os resultados obtidos nesta pesquisa corroboram o uso popular da *J. mollissima* e seu potencial terapêutico.
Palavras-chave: Semi-árido brasileiro. Atividades biológicas. Euphorbiaceae. Plantas medicinais.

Introduction

The use of plants for therapeutic purposes is an ancient practice that perpetuates due to its replication in popular culture. They are results of everyday experiences that had some applicability in the treatment of highly diverse diseases in men or animals and have been transmitted through the generations (ARAÚJO; LEMOS, 2015; SILVA, 2017).

There is no scientific evidence to prove the efficacy of many preparations obtained from the plants or parts of the plants, nor did they undergo any toxicity tests before their administration; their use is strictly based on empiricism. However, these plants present a high level of employability in the primary health care of developing countries, especially where access to health care is scarce (ARAÚJO; LEMOS, 2015; BRAQUEHAIS et al., 2016). Some of these plants arouse a progressive interest in the scientific community, especially in the medical area, due to the antioxidant, antineoplastic, antimicrobial, anti-inflammatory and cicatrizant activities present in organic compounds such as flavonoids, alkaloids, terpenes, tannins, and lignans (ARAÚJO et al., 2008; DEVAPPA et al., 2011; PIMENTEL et al., 2012; SABANDAR et al., 2013; SILVA et al., 2014; ZHANG et al., 2009).

One of these plants is *J. mollissima*, a shrub found in the Northeast, endemic to the caatinga vegetation, which belongs to the family Euphorbiaceae and to the genus *Jatropha*. This genus has many species with various utilities ranging from fuel production and ornamentation, even for medicinal purposes (LEAL; AGRA, 2005; MARIZ et al., 2010; SILVA et al., 2014). In popular medicine, plants of this genus are attributed to various therapeutic properties, such as hemorrhages, ulcers, rheumatism, venereal diseases and purgatives (SABANDAR et al., 2013), with Jatropha (*Jatropha gossypiifolia*) being one of the most studied species. Even so, little is known about the true therapeutic potential of this genus (ROCHA; DANTAS, 2009).

When injured, *J. mollissima* produces a milky-looking sap (latex) that is taken from nature and used by the population on wounds for the purpose of controlling hemorrhages and infections, minimizing the inflammatory process and thus promoting healing. However, this procedure does not have any scientific value because there are few studies about this species related to its use as a hemostatic, antimicrobial, anti-inflammatory and cicatrizant agent, as well as its potential toxic effect (PIMENTEL et al., 2012).

In view of the above, the objective of the present study was to evaluate the chemical composition and pharmacological activities of latex extracts of *J. mollissima* to prove its popular use as a medicinal plant, associating its possible therapeutic effects to the secondary metabolites proposed.

Materials and Methods

Collection and storage of samples

For this study, branches were pruned to collect the latex of the *J. mollissima*. The material was collected in sterile test tubes, wrapped in aluminum foil and sent to the Post-Harvest Laboratory of the Federal Rural Semi-Arid University (UFERSA), where they were kept under refrigeration at 10°C for a maximum of 48 hours until further preparation of the extracts.

Preparation of lyophilized crude latex

Ten milliliters of the crude latex were frozen in a standard freezer at –18°C for 24 h with subsequent removal of the water by sublimation (lyophilization), then it was submitted to vacuum, resulting in the lyophilized crude latex.

Preparation of the aqueous extract (EA)

Dilution of 10 mL of latex in 40 mL of double distilled water was performed by stirring for 24 h. The suspension was then filtered on qualitative filter paper under a vacuum for separation of the water-insoluble material. The resulting solution was frozen in a standard freezer at –18°C for 24 h and then lyophilized to obtain the aqueous extract.

Preparation of hydroalcoholic extract (HAE)

Ten milliliters of the latex in 40 mL of a solution of double distilled water and absolute ethyl alcohol (50% / 50%) was added and stirred up for 24 h. The suspension was then vacuum filtered with qualitative filter paper to remove the insoluble material**,** and the soluble part was concentrated in a rotary evaporator under reduced pressure to remove the ethanol. The remaining aqueous phase was frozen in a standard freezer at –18°C for 24 h and finally freeze-dried.

Preparation of ethanolic extract (EE)

The extract was prepared by adding 10 mL of the latex to 40 mL of absolute ethyl alcohol and stirred up for 24 h. The suspension was then vacuum filtered with qualitative filter paper to separate the insoluble material from ethanol, and the soluble part was concentrated in a rotary evaporator under reduced pressure to remove the ethanol. The remaining product was frozen in a standard freezer at –18°C for 24 h and finally lyophilized.

Animal experimentation and bioethics

Adult female Wistar rats weighing between 180 g and 230 g were used, from the State University of Rio Grande do Norte (UERN) animal facility, where they were kept for adaptation and during the experiment period. They were weighed on a precision digital scale and housed in polypropylene cages (40 x 50 x 20 cm), kept in a room with temperature between (23 to 26°C), controlled relative humidity and light cycle of 12/12 h. Water and feed were offered ad libitum throughout the experiment.

The research protocol was submitted to the Committee of Ethics in Animal Experimental (CEEA) of the State University of Rio Grande do Norte (UERN), protocol number 012/17 and was conducted according to the animal welfare standards in force, according to its council (CONCEA) and to the rules of animal experimentation (Brazilian Federal law nº 11.794/2008).

Histopathological analysis

For this study, were collected fragments of the heart, liver and kidneys were performed and fixed in 10% formaldehyde. After fixation, the fragment organs were submitted to dehydration in increasing concentrations of ethyl alcohol, diaphanized in xylol, and embedded in paraffin to make cutting in microtomes at 5µm, which were stained by hematoxylin and eosin (HE), according to described by Tolosa et al. (2003).

Phytochemical tests

The qualitative phytochemical tests were carried out from chemical reactions that denote the presence or absence of secondary metabolites such as phenols, flavonoids, saponins, anthocyanins, steroids and triterpenoids through alterations in the visual color scale according to the methodology proposed by Matos (2009), while quantification of the total phenolics was done by spectrophotometry, using the Folin-Ciocalteu reagent as described by Hatami et al. (2014).

The total phenol content was determined by interpolating the absorbance of the samples against a calibration curve constructed with gallic acid standards (20 to 200 mg.L $^{-1}$) and expressed as mg EAG (gallic acid equivalent). The equation of the gallic acid calibration curve was y = 0.0096x - 0.0572, where "y" is the concentration of gallic acid, "x" is the absorbance at 760 nm and the correction coefficient R = 0.9991, all analyzes being performed in triplicate.

Assessment of acute toxicity

Eight female Wistar rats were separated into two groups of four animals each. G1 animals were weighed and received a single dose of oral latex, equivalent to 2000.0 mg per kilogram of animal body weight (OECD, 2001). After the administration, the group receiving the treatment was observed for 4 consecutive hours and every 24 h for a period of 14 days, observing the appearance of general toxic signs based on the methodology adapted from Mariz et al. (2006).

During this period of observation, the weight evolution, water and food consumption were measured. The water and feed intake were evaluated daily and counted per group, recording the daily mean values. The body mass was verified at the beginning (d0), middle (d7) and end of the experiment (d14).

At the end of the experiment, the animals were anesthetized with a combination of 2% xylazine hydrochloride (10.0 mg kg $^{-1}$) and 10% ketamine hydrochloride (100.0 mg kg $^{-1}$) intramuscular, and blood was collected through the vein aorta for the hematological and biochemical examinations; the animals were then euthanized by cervical dislocation, as described by Neves (2013). The resection, weighing and macroscopic analysis of the heart, liver and kidneys were performed. Group G2 (control) was submitted to the same procedures, receiving oral distilled water instead of latex.

Evaluation of antioxidant activity

For this procedure, the *in vitro* photocolorimetric method of free radical DPPH (2,2-diphenyl-1-picrylhydrazyl) was used, as described by Mensor et al. (2001). Samples of lyophilized aqueous, hydroalcoholic, ethanolic and crude latex extracts were diluted at concentrations of 1000.0, 750.0, 500.0 and 250.0 ppm, respectively. Then, 1 mL of each sample was mixed with 1.5 mL of DPPH solution (0.02 mg mL $^{-1}$) diluted in methanol. The mixtures were maintained for 15 minutes at 25°C, and in the dark, the absorbances were measured in at 517 nm (spectrophotometer model UV-340G, Gehaka). For the control group, 1 mL of methanol was mixed with 1.5 mL of the DPPH solution. The tests were performed in triplicate**,** and the IC_{50} was obtained from the percent inhibition versus concentration plot, with IC_{50} being the concentration that inhibited the oxidizing power of DPPH by 50%.

Evaluation of antibacterial activity

The bacterial strains used were from standardized collections of the American Type Cell Culture (ATCC), properly characterized morphologically, physiologically and biochemically with Gram-positive bacteria (*Staphylococcus aureus ATCC 25923, Streptococcus agalactiae ATCC 13813,*

Streptococcus pyogenes ATCC 19615, Streptococcus mutans ATCC 25175 and Enterococcus faecalis ATCC 29212) and Gram-negative bacteria (*Shigella flexneri ATCC 12022, Escherichia coli ATCC 25922, Klebsiella pneumoniae ATCC 700603, Salmonella typhimurium ATCC 14028 and Pseudomonas aeruginosa ATCC 27853*).

Initially, all strains were reactivated in brain heart infusion (BHI) for 24 h to obtain turbidity equivalent to the standard 0.5 of the McFarland scale, corresponding to a concentration of approximately 1.5 x 10^8 CFU mL $^{-1}$ (CLSI, 2015). By means of an antibiogram performed in triplicate by the agar diffusion technique (AGID), levofloxacin (5 µg) was shown to have a greater effect on the inhibition of bacterial growth for the strains tested, being used as a positive control; 40 µL of distilled water was used as a negative control.

Determination of minimum inhibitory concentration

The evaluation of the antibacterial activity was verified by the determination of the minimum inhibitory concentration (MIC), which corresponds to the lower concentration of extracts and the latex of *J. mollissima* able to inhibit the abovementioned bacteria. MIC was determined by 96–well plate microdilution tests according to standards established by the Clinical and Laboratory Standards Institute (CLSI, 2015). A total of 20 µL of the McFarland-adjusted inoculum, 100 µL of the Mueller-Hinton agar and 100 µL of the pure latex or extracts thereof were added to each well at the 75%, 50% and 25% dilutions of the parent samples, respectively. In the negative control wells**,** the culture medium, the microorganism inoculum and distilled water were added, while for the positive control**,** the culture medium, inoculum of the microorganism and the antibiotic levofloxacin were added.

After the incubation period at 37°C for 24 h, 10 µl of resazurin (1-hydroxy-3H-phenoxazin-3-one-10-oxide) was added to each well, and the microplates were again incubated at 37°C for 2 h.

During the plates reading, after the incubation time, a change of the blue color (resazurin original) to pink was observed, characterizing the reduction of this dye where there was bacterial viability; in the wells where the color remained blue, there was no reduction of the dye, indicating bacterial unviability and, consequently, inhibition of microorganisms (GONÇALVES et al., 2012). All the tests were performed in triplicate.

Determination of minimum bactericidal concentration

To evaluate the bactericidal and/or bacteriostatic potential of the latex and its extracts, 10 µL from the wells used in the MIC tests, where there was no visible bacterial growth, were removed and sowed on the surface of a Mueller-Hinton agar plate. After 48 h of incubation at 37ºC, the minimum bactericidal concentration (MBC) was defined as the lowest concentration of latex and extracts under study capable of causing death of the microorganisms, as described by Santurio et al. (2007).

Statistical analysis

The quantitative data were analyzed using spreadsheets and by applying descriptive statistical techniques, and the data were represented by the mean and standard deviation. Comparisons between groups were assessed using Student's t-test or analysis of variance (ANOVA). Subsequently, we performed multiple post-hoc Tukey's t tests when the results were considered normal, and the Kruskal-Wallis test when the results were abnormal, both followed by the Mann-Whitney test; significant values were those of $p \leq 0.05$.

Results and Discussion

Phytochemical tests

The prospects of the secondary compounds present in the extracts of *J. mollissima* and in plants of the genus have already been previously described

in some studies (ARAÚJO et al., 2008; DEVAPPA et al., 2011; GOMES et al., 2016; SABANDAR et al., 2013; SILVA et al., 2014; ZHANG et al., 2009). However, the extraction of these constituents is directly related to the method, the solvents and even the parts of the plants used in the preparation, allowing variations in the same specimen and justifying their evaluation in the extracts under study. In the present study, the prospecting of the chemical constituents of extracts of *J. mollissima* showed positive results for phenolic compounds, such as tannins, flavonoids and saponins (Table 1).

In a study with 33 species of plants of the caatinga, chosen based on the popular belief for supposed healing and anti-inflammatory effects, Araújo et al. (2008) inferred that these effects were derived from phenolic compounds, such as tannins and flavonoids, found in the latex of *J. mollissima* and other plant species. The anti-inflammatory effects of these plants were associated with tannin, but this relationship was not associated with the presence of flavonoids. Melo et al. (2010) investigated the tannin contents and antioxidant and antineoplastic properties of extracts from 14 plants from the semi-arid region, detecting high levels of tannins and the antioxidant power of *J. mollissima*, which corroborates the findings of our study.

Gomes et al. (2016) investigated the antiofidic effect of the aqueous extract of *J. mollissima* leaves. Through the analysis of thin layer chromatography, it was possible to conclude that flavonoid and saponin compounds predominate in their constitution. Braquehais et al. (2016) performed a phytochemical analysis of the ethanolic extract of the leaves of *J. mollissima* and verified the presence of coumarins, tannins, flavonoids, alkaloids and steroids but did not detect the presence of triterpenoids, which is also in line with the results of this research.

Table 1. Qualitative analysis of secondary metabolites of *J. mollissima* from visual color scale or foaming. Aquareous Extract (AE), Hydroalcoholic Extract (HAE), Ethanolic Extract (EE).

Bioactive compounds	Latex	AE	HAE	EE
Phenols	+++	+	++	+++
Tannins	+++	+	++	+++
Flavonoids	+++	+	++	+++
Saponins	+++	+	++	+++
Steroids	–	–	–	–
Triterpenoids	–	–	–	–
Anthocyanins	–	–	–	–

(–) Absence of color or foaming; (+) Poor color or formation of low foam; (++) Attenuated color or moderate foam formation; (+++) Intense color or abundant foam formation.

Review studies on the medicinal and phytochemical properties of *Jatropha* plants identified alkaloids, terpenes, flavonoids, cyclic peptides, lignans, phenolic compounds, saponins, steroids, among others, in various parts of plants such as latex cyclic peptides and terpenes in roots and stems (DEVAPPA et al., 2011; SILVA et al., 2014; ZHANG et al., 2009).

The quantification of the total phenols in the latex and its extracts, obtained through a calibration curve with a standard of equivalence to gallic acid, allows us to infer that all had low levels of phenolic compounds (Table 2), ranging from 0.82 mg EAG g^{-1} present in the aqueous extract at 58.07 mg EAG g^{-1} in the latex.

Table 2. Quantitative analysis of total phenolics, represented as the mean ± standard deviation of the mean. Statistical difference calculated using ANOVA associated with Tukey's t test ($p \leq 0.05$).

Sample	mgEAG.g $^{-1}$
Aqueous Extract	0.82 ± 0.02 a
Hydroalcoholic Extract	45.68 ± 0.43 b
Ethanolic Extract	50.00 ± 0.97 b
Latex *in nature*	58.07 ± 0.77 b

Different letters indicate significant differences.

Evaluation of toxicity

There were no deaths among G1 rats given the dose of 2000.0 mg kg $^{-1}$ of latex, which suggests low toxicity and safety in its administration, in view of other studies using this same dose.

The behavioral changes observed after administration of the latex were initial agitation, followed by lethargy and tachypnea, which disappear within a few hours. Motor and/or sensory changes were not observed or had little relevance when compared to their controls.

The average daily water consumption was 107.0 mL for G1 and 120.0 mL for G2 (control). For the average daily feed intake, there was an average intake of 47.4 g and 64.1 g daily for G1 and G2 (control), respectively. These differences occurred mainly in the first two days of the study, in which G1 animals presented hyporexia and oligodipsia, equating consumption to G2 (control) on subsequent days, without significant difference between the groups.

Among hematological and biochemical parameters, only hemoglobin and mean corpuscular hemoglobin (MCHC) showed a significant reduction in the group treated with latex, when compared to the control group. However, the cell lines (red blood cells, leukocytes and platelets) did not show significant changes, suggesting absence or low medullary toxicity, at the dose adopted. Functional evidence and markers of liver injury (AST, ALT, GGT and albumin), as well as renal function (urea and creatinine), did not show any significant differences compared to G2 (control), evidencing the absence or low hepatic and renal toxicities in the same doses. These results, except for hemoglobin, leukocyte and platelet counts, are similar to those obtained from the central laboratory of the Federal University of Sergipe (MELO et al., 2012).

G1 animals in the first week of the experiment had a reduction in body mass and almost complete recovery at term (d14). G2 (control) presented a linear growth of the masses at the end of the test (d14), but these changes were not significant ($p \leq 0.05$). The organs, liver, kidneys and heart showed no macroscopic and microscopic changes, nor did their respective masses present significant differences, in relation to the control group ($p \leq 0.05$).

According to Ribeiro et al. (2014), the ethanolic extract obtained from the *J. mollissima* stem was toxic in the medium lethal concentration test (LC_{50}) for *Artemia salina*, reaching 660.80 μg mL $^{-1}$, while the ethanolic extract obtained from the leaves, according to Braquehais et al. (2016), presented an LC_{50} equal to 406.02 μg mL $^{-1}$ for this same test. Idealized by Meyer et al. (1982), this method considers toxic extracts with LC_{50} less than 1000 μg mL $^{-1}$.

Mariz et al. (2006) obtained an ethanolic extract derived from the leaves and stem of *J. gossypiifolia* and tested it in Wistar rats at doses up to 5000.0 mg kg $^{-1}$. This test showed low acute oral toxicity at doses below 2000.0 mg kg $^{-1}$ of these extracts and minor histopathological changes in the animals treated with the highest experimental dose (MARIZ et al., 2008), which was similar to the results obtained in this study.

Several studies have demonstrated the therapeutic effects of plant extracts; however, such products should be evaluated through toxicological tests capable of establishing safety intervals in use, as well as side effects and adverse reactions. In this study, the value of LC_{50} for the oral administration of *J. mollissima* latex was higher than 2000.0 mg kg $^{-1}$, which falls into category 5 (low toxicity) according to the Globally Harmonized System.

Evaluation of antioxidant activity

The antioxidant activity was evaluated by the determination of DPPH (IC_{50}) radical uptake by *J. mollissima* latex and its extracts. The lower the IC_{50} value, the higher the antioxidant potency, since a smaller amount of the substance will be required to promote a 50% reduction of the reagent. In this context, the latex was more effective when compared to the ethanolic extract, which in turn proved to be more effective than the hydroalcoholic extract, with both being more effective than the aqueous extract, which may be related to the high presence of phenolic compounds (Table 3).

Table 3. Antioxidant activity is represented as the mean ± standard deviation. Significant differences were calculated using ANOVA, associated with Tukey's t test ($p \leq 0.05$).

Sample	IC_{50} (ppm)
Aqueous Extract	17759.83 ± 3267.63 a
Hydroalcoholic Extract	167.66 ± 7.04 b
Ethanolic Extract	127.30 ± 3.90 b
Latex *in natura*	124.27 ± 7.75 b

Different letters indicate significant differences.

Melo et al. (2010) included *Poincianella pyramidalis*, *Jatropha mollissima*, *Anadenanthera colubrina* and *Croton blanchetianus* in their work, because they show a lower IC of 50. The control used in this assay was ascorbic acid, which showed an IC_{50} of 21.74 ± 3.23 µg. In general, the IC_{50} values of the plants analyzed ranged from 42.95 to 94.41 ± 2.67 µg mL^{-1}, with an IC_{50} of *J. mollissima* of 54.09 ± 4.36 µg.mL^{-1}.

Melo et al. (2010) classified the antioxidant activity of its extracts as: good (IC_{50} < 65.0 µg), moderate (65.0 µg < IC_{50} < 152.0 µg) and low (IC_{50} > 152.0 µg) activity. In this case, it is concluded that *J. mollissima* showed a good antioxidant activity, which showed moderate action for latex and ethanolic extract, and low for hydroalcoholic and aqueous extracts. This fact may be associated to the extraction method, since different solvents were used, or the parts of the plant used in the preparation.

Antioxidant activity is closely related to phenol content because phenolic compounds are included in the classification of antioxidants (MELO et al., 2010). Thus, we can observe that this relationship was proven for the latex of *J. mollissima*, because it presented better antioxidant activity and a higher content of total phenols. In contrast, the aqueous extract had the lowest value for total phenols and the lowest antioxidant activity.

Evaluation of antibacterial activity

The results obtained, according to variations in the latex or extracts concentration, are shown in Table 4. The change from blue to pink color is symbolized by (–), which characterizes the reduction of resazurin dye in wells where there was bacterial viability. The wells where the color remained blue are indicated by (+), as there was no reduction of

the dye, indicating bacterial non-viability and, consequently, inhibition of the microorganisms. The MBC test verified that there was no bacterial death and the extracts analyzed were bacteriostatic.

The antibacterial activity of latex and its extracts may be related to the action of phenolic compounds, markedly tannins and flavonoids present in their compositions. These secondary metabolites can act by causing hydrolysis of ligands or by forming complexes with bacterial proteins, promoting the breakdown of the plasma membrane and cell wall of the bacterium, resulting in its destruction (BRAQUEHAIS et al., 2016).

Because they are large molecules of low polarity, it is believed that these secondary compounds were not extracted in the preparation of the aqueous extract, which would justify it being the only extract with no effect at any of the concentrations tested. The opposite is true because the presence of alcohol, the most apolar substance, enabled the extraction of these metabolites, providing a greater antibacterial effect in the other extracts.

Table 4. Determination of minimal inhibitory concentration ($mg.mL^{-1}$) in resazurin assay 0.01%.

Samples / Strains	Latex				Ethanol extract				Hydroalcoholic extract				Aqueous extract			
	100	75	50	25	100	75	50	25	100	75	50	25	100	75	50	25
P. aeruginosa	+	+	–	–	+	+	+	+	+	+	+	+	–	–	–	–
S. typhimurium	+	+	–	–	+	+	+	+	+	+	–	–	–	–	–	–
K. pneumoniae	+	+	+	+	+	+	+	+	+	+	+	+	–	–	–	–
E. coli	+	+	–	–	+	+	+	+	+	+	+	+	–	–	–	–
S. flexneri	+	+	+	–	+	+	+	+	+	+	+	+	–	–	–	–
E. faecalis	+	+	+	–	+	+	+	+	+	+	+	+	–	–	–	–
S. agalactiae	+	+	+	–	+	+	+	+	+	+	+	+	–	–	–	–
S. pyogenes	+	+	+	–	+	+	+	+	+	+	+	+	–	–	–	–
S. mutans	+	+	–	–	+	+	+	+	+	+	+	+	–	–	–	–
S. aureus	+	+	–	–	+	+	+	+	+	+	+	+	–	–	–	–

Camelo (2015) found an MIC of 8192.0 μg mL^{-1} for Gram-positive *S. aureus*, *E. faecalis* and Gram-negative bacteria *E. coli* and *P. aeruginosa* using *J. mollissima* latex. Rocha and Dantas (2009), in a comparative study of MICs between the latex of *J. mollissima* and *J. gossypiifolia*, found that both plants presented antibacterial activity against *S. aureus* and *S. typhimurium*.

Silva (2017), studying the antimicrobial activity of the leaves, branches and stem of *J. gossypiifolia* against *S. aureus*, *P. aeruginosa* and *E. coli* strains, found the smallest MIC's, or the highest antibacterial activity, in the leaves against *S. aureus* (500.0 μg mL^{-1}), leaves and branches against *P. aeruginosa* (500.0 μg mL^{-1}), but found no activity against *E. coli*. These findings were partially at odds with the results of our study and of Camelo (2015), which also showed activity of latex and EE and HAE against *E. coli*. In the *in vitro* evaluation, Braquehais et al. (2016) found antibacterial action in the ethanolic extract of the leaves of *Jatropha mollissima* against the strains *E. coli, P. aeruginosa, K. pneumoniae* and *E. faecalis* only at a concentration of 250.0 mg mL^{-1}.

To delimit the antibacterial potential of extracts derived from plants, it was determined that MIC values below 100.0 μg mL^{-1} had good antimicrobial activity, values between 100.0 and 500.0 μg mL^{-1} were moderately active and values between 500.0 and 1000.0 μg mL^{-1} and MIC greater than 1000.0 μg mL^{-1} were inactive (HOLETZ et al., 2002).

According to the results obtained, it can be concluded that although our research was qualitative, the findings suggest that *J. mollissima* latex and ethanolic and hydroalcoholic extracts can be used as antibacterial agents because, even when very diluted, they were able to inhibit the strains tested.

The latex and ethanolic extracts of *J. mollissima* presented expressive amounts of bioactive compounds, such as tannins, flavonoids, and saponins, in addition to moderate antioxidant activity. For the administered dose, latex also had low acute toxicity in Wistar rats.

The *in vitro* antibacterial activity assays showed that even at low concentrations, latex and its extracts, both ethanolic and hydroalcoholic, promoted growth inhibition of both the gram (+) and gram (–) bacteria tested for some strains.

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