Fronteiras: Journal of Social, Technological and Environmental Science
http://periodicos.unievangelica.edu.br/fronteiras/
ISSN 2238-8869

Article

Comparative Study of Commercial Saps of *Croton lechleri*: Organoleptic, Raman Spectroscopy, Phytochemical Prospecting and Acute Toxicity Analysis

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ABSTRACT
Dragon's blood resin oil from the Amazon has multivariate ethnopharmacological potential, but there is a lack of scientific evidence regarding the quality control of this natural product. The objective of this study was to compare different commercial saps regarding their organoleptic, physicochemical, phytochemical, and toxicological characteristics. Nine commercial samples of *Croton lechleri* were tested for organoleptic properties, Raman spectroscopy, phytochemical screening, and toxicological analysis using the *Artemia salina* model. The *C. lechleri* saps showed sensory differences with a higher prevalence of burgundy color, soapy effect, fluidity, and woody odor, and few of them exhibited small residues. There was a general tendency towards moderate spreading and density and intense dehydration. Similar band spectra and vibrational intensities suggested the presence of alkaloids such as taspine, as well as terpenes, catechins, tannins, and polyphenols. The vast majority showed peaks consistent with condensed tannins, chalcones, aurones, flavonols, flavanones, flavanonols, xanthones, catechins, steroids, diterpenes, saponins, and alkaloids. The acute toxicity test with microcrustaceans showed an LC50 greater than 1000 µg/mL, significantly different from potassium dichromate. Continued preclinical research with dragon's blood sap is needed to validate its safe and effective use.
Key words: dragon's blood; *Croton lechleri*; bioprospecting; ecotoxicology; phytotherapeutic drugs.

RESUMO
O óleo de resina de sangue de dragão da Amazônia possui potencial etnofarmacológico multivariado, porém há carência de evidências científicas sobre o controle de qualidade deste produto natural. O objetivo deste estudo foi comparar diferentes seivas comerciais quanto às suas características organolépticas, físico-químicas, fitoquímicas e toxicológicas. Nove amostras comerciais de *Croton lechleri* foram testadas quanto às propriedades organolépticas, espectroscopia Raman, prospecção fitoquímica e análise toxicológica utilizando o modelo de *Artemia salina*. As seivas de *C. lechleri* apresentaram diferenças sensoriais com maior prevalência de cor bordô, efeito ensaboado, fluidez e odor amadeirado, e poucas delas exibiram pequenos resíduos. Houve tendência geral de espalhamento e densidade moderados e desidratação intensa. Espectros semelhantes de bandas e intensidades vibracionais sugeriram a presença de alcaloides como taspina, além de terpenos, catequinas, taninos e polifenóis. A grande maioria apresentou picos consistentes com taninos condensados, chalconas, auronas, flavonóis, flavanonas, flavanonóis, xantonas, catequinas, esteroides, diterpenos, saponinas e alcaloides. O teste de toxicidade aguda com microcrustáceos foi verificada por CL50 superior a 1000 µg/mL, significativamente diferente do dicromato de potássio. Há necessidade de pesquisas pré-clínicas contínuas com seivas de sangue-de-dragão para validar seu uso seguro e eficaz.
Palavras-chave: sangue de dragão; *Croton lechleri*; bioprospecção; ecotoxicologia; medicamento fitoterápico.

Submissão: 14/06/2025 Aceite: 25/05/2026 Publicação: 14/11/2026

v.15, n.3, 2026 • p. 53-69. • DOI http://dx.doi.org/10.21664/2238-8869.2026v15i3.8120

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Introduction

Traditional medicine holds a significant place within the Brazilian cultural repertoire, rooted in the empirical practices of vulnerable communities lacking formal health care services, and further developed through the ethnopharmacological perspective based on scientific investigations of plant bioactives with therapeutic potential. The Amazon biome represents a fertile ground for rational biotechnological exploration, which must be aligned with the preservation of natural resources amid unprecedented environmental devastation and global climate urgency, as well as with the equitable distribution of associated benefits (Lopes et al. 2013; Kellner 2021).

Investment in basic science to understand biodiversity and its medicinal chemistry is essential for the discovery of new natural products, with the potential to generate billions of US dollars in economic value (Kellner 2021). Brazil stands out in scientific production leveraging Amazonian resources for the development of phytomedicines, particularly for neglected diseases; however, technological maturity remains at early laboratory stages compared to the USA, where clinical investigation is more advanced. Therefore, expanding collaborative research networks is necessary to translate such resources into global health benefits (Lepsch-Cunha et al. 2024).

Dragon's blood is a popular name given to the dark red natural resin employed by many cultures since the medieval era for artistic, medical, alchemical, or mystical purposes. It is obtained from various plant species, including *Dracaena cinnabari* (Convallariaceae) on the island of Socotra, *Dracaena draco* in the Canary and Madeira Islands, and species of the genera Daemonorops (Palmae) in Southeast Asia and Croton (Euphorbiaceae) in tropical regions of Africa, Asia, and the Americas (Edwards et al. 2004).

In particular, *Croton lechleri Müll. Arg.,* with botanical synonyms *Croton draco var. cordatus Müll. Arg.* and *Oxydectes lechleri Kuntze*, is a tree native to the Amazon rainforest of South America, with confirmed occurrence in Peru (Cayo et al. 2021), Brazil (Martins et al. 2016; Perín et al. 2024; Silva et al. 2022), Ecuador, Colombia, and Bolivia, and also been reported in Mexico (Buitrón & Gonzales 2019; Hidalgo et al. 2018; Londoño-Lemos & Bustamante 2019; Rodríguez-Garcia et al. 2010). The taxonomic identification of *C. lechleri* is complex, given the considerable morphological variation in terms of shape, arrangement, consistency, color and size of leaves, trichomes and glands, age, geographic distribution and adaptive processes (Lopes et al. 2013).

The pure sap of *C. lechleri* is rich in secondary metabolites, including alkaloids such as taspine (Monterubbianesi et al. 2023), protoanthocyanidins, and aporphine, polyphenols such as flavonoids, catechins, and 3'-4-*O*-dimethylcedrusine (Martins et al. 2016; Perín et al. 2024); terpenes (Silva et al. 2022), as well as pyridone, quinoline, phytosterols (Rodríguez-Garcia et al. 2010), leptins, acetylsalicylic acid, and anthraquinones (Londoño-Lemos & Bustamante 2019; Hidalgo et al. 2018; Cayo & Barrera 2014; Monterubbianesi et al. 2023).

Such phytochemicals are associated with multiple medicinal properties. Antimicrobial activities include antibacterial (Cayo et al. 2021), antiviral, antifungal (Monterubbianesi et al. 2023), and antiparasitic effects (Correa-Olaya et al. 2021; Silva et al. 2022). Anti-inflammatory, analgesic, and wound healing properties have also been reported (Cavalie-Martel et al. 2019), alongside antioxidant (Perín et al. 2024) and antitumor activities (Silva et al. 2022). Additionally, thrombolytic and antiplatelet effects have been described, with no evidence of hemolytic activity (Cayo et al. 2021; Cayo & Barrera 2014; Correa-Olaya et al. 2021; Londoño-Lemos & Bustamante 2019; Hidalgo et al. 2018; Monterubbianesi et al. 2023; Rodríguez-Garcia et al. 2010; Silva et al. 2022).). Such biological properties have supported clinical and preclinical investigations of *C. lechleri* across multiple therapeutic areas, particularly in medicine and dentistry.

In medicine, topical or enteral applications of *C. lechleri* have been reported for antiulcer, antidiarrheal, anticancer, and antitumor purposes, including treatment of carcinomas, melanomas, and gynecological conditions (Lopes et al. 2013). In dentistry, the sap has demonstrated a broad in vitro antimicrobial spectrum against cariogenic (Hidalgo et al. 2018; Cayo & Barrera 2014) and periodontopathogenic strains (Rodríguez-Garcia et al. 2010), as well as antimicrobial effects when applied by brushing or rinsing (Londoño-Lemos & Bustamante 2019).

Furthermore, at unidentified concentrations (Londoño-Lemos & Bustamante 2019) or ranging from 2.5% (Correa-Olaya et al. 2021) to 10% (Martins et al. 2016), *C. lechleri* sap has demonstrated analgesic, anti-inflammatory, and repair effects in both preclinical studies, such as in rabbit dental pulp (Correa-Olaya et al. 2021), and human treatments, including application to the periodontal ligament after tooth avulsion (Martins et al. 2016), dental alveoli after tooth extraction (Cavalie-Martel et al. 2019), dentin-pulp complex after removal of carious tissue (Londoño-Lemos & Bustamante 2019), and oral mucosa with recurrent aphthous stomatitis and gingival cleft (Buitrón & Gonzales 2019; Monterubbianesi et al. 2023). From an economic standpoint, *C. lechleri* sap represents a natural, accessible, and low-cost alternative (Hidalgo et al. 2018), with potential to benefit

small producer communities in Amazonian regions and the national pharmaceutical industry in search of therapeutic bioactives (Lopes et al. 2013).

The few recent studies on dragon's blood sap of Brazilian origin, all conducted in the Northern Region of Brazil, from the states of Rondônia (Martins et al. 2016; Perín et al. 2024) and Amazonas (Silva et al. 2022), have focused less on identifying its active ingredients (Perín et al. 2024) than on understanding its biological effects, related to toxicity (Silva et al. 2022) or tissue repair (Martins et al. 2016).

A search of the Agência Nacional de Vigilância Sanitária database (2025) revealed that products containing dragon's blood, classified as grade 1 cosmetics, exempt from registration but notified to the agency, totaled 49 items with active regulatory status, of which only 13 were concentrated saps or resinous oils in pure or 100% natural form; the vast majority consisted of derivatives with additives, including essential oils, incense, soaps, facial or lip serums, body moisturizers, hair masks, hair styling wax, shampoos, conditioners, and toothpaste.

Although commercial products with different presentations and geographical origins of raw material extraction may differ in chemical composition, structure, and bioactivity, which may interfere with their therapeutic efficacy, comparative data on their organoleptic, physicochemical, phytochemical, and toxicological properties remain scarce in the literature. We hypothesized that commercial saps of *C. lechleri* from different geographical origins and manufacturers would present distinct profiles, reflecting the absence of standardized quality control for these products. Therefore, the present study aimed to characterize and compare different commercial saps of *Croton lechleri*.

Material and Methods

Study design

The methodological typology of this scientific study followed an experimental laboratory modality, with an analytical objective and a quali-quantitative approach.

Ethical aspects

A sap acquired in commerce as ex situ biodiversity and with an unidentifiable origin from a traditional community was registered in the National System of Genetic Heritage and Associated Traditional Knowledge (SISGEN) as ethical-legal registration in the Genetic Heritage System of the Ministry of the Environment in accordance with Law no. 13,123/2015 (Brasil 2015). The use of microcrustaceans in bioassay, as it does not fall into the category of tests with chordate animals, also did not require bioethical assessment for its management and use, in accordance with Law no. 11,794/2008 (Brasil 2008). As the object of the research did not involve therapeutic application in human beings, there was no need for assessment by an ethics committee, as provided for in Law no. 14,874/2024 (Brasil 2024).

Samples

A sample of dragon's blood sap was selected from a popular market in the city of Altamira, Pará, Brazil (G1). The selection of manufactured test products was based on a random sample, guided by marketing criteria, such as: registered or well-known company brands, electronic advertising and marketing, affordable price, and product availability (G2-G9). Commercial samples of dragon's blood, self-referenced on the packaging or labeling as consisting entirely of pure sap or undiluted oleoresin of *Croton lechleri*, are specified in Table 1.

Table 1. Description of experimental groups of dragon's blood sap from *Croton lechleri*.

Group	Product
G1	Dragon's blood sap, Ponto das Ervas - Box no. 112/Municipal Market (Altamira, PA, Brazil). SISGEN authorization no.: A0025B1/2022. No traceable information at ANVISA. Manufacture and expiration date: undetermined.
G2	Dragon's blood sap, Phytoterapica™ (Guarulhos, SP, Brazil). Batch: PSD223. ANVISA authorization no.: 2.03883-7, process no. 25351.646453/2013-51. Manufacture: undetermined. Expiration date: Nov. 2024.
G3	Dragon's blood resinous sap, Terra Flor™ (Alto Paraíso de Goiás, GO, Brazil). Batch: 22190. ANVISA authorization no.: 2.09199-3, process no. 25351.419400/2017-60. Manufacture: undetermined. Expiration date: Sep. 2024.
G4	Dragon's blood essential oil, Herbia™ (Curitiba, PR, Brazil). Batch: 5F3/23. ANVISA authorization no.: 2.03086-4, process no. 25351.623262/2023-92. Manufacture: Mar. 2023. Expiration date: Mar. 2026.
G5	Dragon's blood oleoresin, Laszlo™ (Belo Horizonte, MG, Brazil). Batch: LZ2069. ANVISA authorization no.: 2.03740-2, process no. 25351.450246/2020-21. Manufacture: undetermined. Expiration date: Nov. 2026.
G6	Dragon's blood resinous sap-essential oil, Bauru Distillery (Catanduva, SP, Brazil). Batch: 23112. No traceable information at ANVISA. Manufacture: Mar. 2023. Expiration date: Mar. 2025.
G7	Dragon's blood sap vegetable oil, Artesanal (Vale do Jamari, RO, Brazil). Batch: 22190. No traceable information at ANVISA. Manufacture: Sep. 2023. Expiration date: Sep. 2025.
G8	Dragon's blood oil, Saúde da Terra (Santa Teresinha, BA, Brazil). Batch: 02/22. No traceable information at ANVISA. Manufacture: Feb. 2022. Expiration date: Feb. 2025.
G9	Dragon's blood/Sangre de grado, Amazonas (Amazon, Peru). Batch: LC035/20. No traceable information at ANVISA. Manufactured: Sep. 2022. Expiration date: Sep. 2025

Source: Research data.

Although this sampling approach does not follow a probabilistic design, it was intentionally adopted to reflect the true diversity of commercial dragon's blood products available on the Brazilian market, since differences in composition may reflect, in part, the nature of each product type, and not just geographical or manufacturer-related variations.

Most samples (G2, G3, G4, G5, G6 and G7) remained in amber glass bottles while the others (G1, G8 and G9) were in opaque plastic bottles. The information regarding the validity of the sap was present in most samples (G2 to G9), except in G1, without data on the label. All were marketed and stored in the laboratory at room temperature until the time of testing.

Organoleptic analysis

The organoleptic qualities, detectable by the sense organs, were assessed with the naked eye under direct lighting and classified in agreement by two previously calibrated examiners. Visual characteristics included color, in the reddish brown or burgundy, dark brown, light brown or amber and colorless patterns, and the presence of small residues in each original sample. The soapy effect, whether present or not, and the fluid or sticky viscosity were assessed by tactile contact. The typical woody/resinous, fragrant or fruity odor was detected by direct olfaction.

Physicochemical analysis

The qualitative and quantitative morphological description of the degree of liquid spreading was performed through representative photographic records of 0.5 mL wet aliquots of the saps at the immediate time (T0) and of the dry or semi-fluid samples at room temperature after 3 h (T3) captured in a trinocular stereomicroscope model DI-106T (Digilab, Sao Paulo, Brazil) attached to a digital camera model NewValue 48 Megapixel (Panasonic, Sao Paulo, Brazil) at 10x magnification. The initial and final areas in mm^2 were outlined and calculated using ImageJ software version 1.54 (National Institutes of Health, USA), which allowed to plot an absolute and relative percentage of dispersion for each sample. Subsequently, the liquid samples at T0 and solid or semi-solid samples were left to dry continuously for 12 h (T12) and weighed on an analytical balance (Shimadzu Corporation, Kyoto, Japan) to evaluate the possible dehydration and loss of mass of the saps. To determine the density (g/mL), considering the initial volume of 0.5 mL, the initial value of the weight of each sample was multiplied by two to obtain the respective indicator.

For spectral analysis, 1 mL of sap was dried at room temperature (27°C) for 12 hours under standard laboratory conditions at the Biomaterials Laboratory of the Federal University of Ceará (UFC), Sobral Campus, without automated drying means to avoid thermal degradation of the bioactive compounds. The samples were crushed and positioned in an Xplora Raman spectrometer (Horiba, Paris, France) containing an argon laser with a wavelength of 532 nm and a power of 10 mW. Objective lenses (Olympus Corporation, Shinjuku, Tokyo, Japan) with 10x and 100x magnification were used to focus on the regions to be evaluated, thus avoiding cracks and residues in the samples that could interfere with the reading. The Raman spectrum was obtained in the range of up to 2000 cm^{-1}, with spectral scans at a resolution of 4 cm^{-1}, with a total of 25 accumulations per region. Data preprocessing included baseline correction of the spectra and was performed through parabolic subtraction to improve the signal-to-noise ratio and spectral analyses, using the Origin® 7.0 program (OriginLab Corporation, Northampton, MA, USA).

Phytochemical analysis

Phytochemical prospecting tests were performed following the methodologies proposed by Matos (2009), Morais and Vieira (2021) and Moreira et al. (2024) which are based on the characteristic colorimetric chemical reaction of secondary metabolite groups, defining the presence of each proposed variable as a positive result and the absence of each proposed variable as a negative result. The only exception is the test for saponins, whose positivity is based on the formation of foam after shaking.

Most tests used alcoholic extracts, obtained by weighing 50 mg of each sample and diluting it in 20 mL of ethanol. Aliquots of 2-3 mL per test tube were used for each qualitative analysis.

In the test for phenols and tannins, a tube with 2 mL of the alcoholic extract had three drops of an alcoholic solution of ferric chloride ($FeCl_3$) added, followed by shaking. Compared with a blank test (water and $FeCl_3$), the blue and red hue would indicate the presence of phenols, blue would indicate hydrolyzable tannins and green would indicate condensed tannins.

In the test for anthocyanins, anthocyanidins, flavones, flavonols, xanthones, chalcones, aurones and flavononols, the first tube with 2 mL of the alcoholic extract was acidified with hydrochloric acid (0.1 M HCl) to pH 3, the second tube was alkalinized with sodium hydroxide (0.1 M NaOH) to pH 8.5 and the third tube was alkalinized to pH 11. In an acidic medium, the red coloration would indicate the presence of anthocyanins, anthocyanidins or chalcones and aurones, while in an alkaline medium the lilac or blue-purple coloration would indicate anthocyanins and anthocyanidins, the yellow coloration would indicate flavones, flavonols and xanthones, the red-purple coloration would indicate chalcones and aurones and the red-orange coloration would indicate flavonols.

In the test for leucoanthocyanidins, catechins and flavanones, the first tube containing 2 mL of the alcoholic extract was acidified with 0.1 M HCl to pH 3 and the second tube was alkalized with 0.1 M NaOH to pH 11, followed by heating in an alcohol lamp for 2 minutes. Compared to the corresponding tubes in the previous test, in the acidulated samples the red coloration would indicate the presence of leucoanthocyanidins and the yellowish-brown coloration would indicate catechins, while in the alkalized samples the red-orange coloration would indicate flavanones.

In the test for flavonols, flavanones, flavanonols and xanthones, a few centigrams of magnesium in strips and 1 mL of concentrated HCI were added to a tube with 2 mL of the alcoholic extract, generating effervescence and at the end of the reaction, in comparison to the acidified tube with pH 3 in the previous test, an intense red color would indicate the presence of flavonols, flavanones, flavanonols and/or xanthones, free ones or their heterosides.

In the test for catechins, 1 mL of a 1% aqueous vanillin solution and 1 mL of HCI P.A. were added to a tube containing 3 mL of the alcoholic extract. The appearance of a red color would indicate the presence of catechins.

In the Liebermann-Burchard test for steroids and triterpenoids, 50 mg of each sample was dissolved in 2 mL of chloroform, with careful trituration of the residues with the solvent, and the chloroform solution was filtered drop by drop into a small funnel closed with a cotton ball and covered with a few decigrams of anhydrous sodium sulfate (Na_2SO_4). 1 mL of acetic anhydride ($[CH_3CO]_2O$) was added to the aliquot in the test tube, gently stirred, three drops of concentrated sulfuric acid (H_2SO_4) were added, and gently stirred again. The rapid development of an evanescent blue color followed by a permanent green color would indicate the presence of free steroids, while a brown to red color would indicate free pentacyclic triterpenoids.

In the test for diterpenes, 3 to 4 drops of 10% copper acetate solution ($Cu[CH_3COO]_2$) were added to a tube containing 2 mL of the alcoholic extract. The emerald green color would indicate the presence of diterpenes.

In the saponin test, 50 mg of each pure sample was added to a tube, diluted in 2 mL of water and followed by vigorous shaking for 2 minutes. The appearance of persistent and abundant foam would indicate the presence of saponin heterosides.

In the test for quinones, 2g of extract and 4mL of ethyl ether or enough to cover the sample were added, the solution was stirred for 2 minutes and filtered into another test tube using cotton. To the new tube, 1mL of 10% NaOH aqueous solution was added. The pinkish-red coloration would indicate the presence of hydroxylated anthraquinones.

In the test for alkaloids, 50 mg of each pure sample was added to two test tubes, which were then acidified with 1 mL of 0.1 M HCl. Three drops of Bouchardat reagent were added to the first tube and three drops of Dragendorff reagent to the second tube. A positive reaction for alkaloids would be indicated in the first tube by the appearance of a reddish-orange precipitate, while in the second tube by a brick-red precipitate.

In the coumarin test, 3 drops of the concentrated extract were placed on a 3x5 cm filter paper strip to form two spots approximately 1.5 cm in diameter each. Spot 1 was the control, and 1 drop of 10% (m/v) NaOH solution was added to spot 2, which was kept at room temperature until dry. The strip was then placed in a chamber with ultraviolet light and observed for 2 to 3 minutes. A yellow or green coloration would indicate the presence of coumarin compounds.

Toxicological analysis

The acute toxicity bioassay with *Artemia salina* was performed in accordance with ISO/TS 20787 (2017). To produce the artificial seawater solution, the minerals sodium chloride (NaCl, 24.53 g/L), magnesium chloride ($MgCl_2$, 5.20 g/L), sodium sulfate (Na_2SO_4, 4.09 g/L), calcium chloride ($CaCl_2$, 1.16 g/L), potassium chloride (KCl, 0.695 g/L), sodium bicarbonate ($NaHCO_3$, 0.201 g/L), potassium bromide (KBr, 0.101 g/L), boric acid (H_3BO_3, 0.027 g/L), strontium chloride ($SrCl_2$, 0.025 g/L), and sodium fluoride (NaF, 0.003 g/L) were diluted in distilled water. The microcrustacean cysts remained for 48h in this saline solution, in light/dark cycles, pH 8.0±0.5 and constant aeration at 25 ºC. After hatching, ten viable, mobile and phototropic nauplii were transferred to each 1 mL well, containing the aqueous extract of the samples at the following concentrations in quintuplicate: 10, 100, 500, 1,000 and 5,000µg/mL. Saline solution was chosen as the negative control and potassium dichromate/$K_2Cr_2O_7$ (Êxodo Científica, Sumaré, SP, Brazil) as the negative control, since a toxic chemical agent was expected, and was placed in the same concentrations as in the tests. After 24 hours of incubation, the nauplii were counted visually using a trinocular stereomicroscope model DI-106T (Digilab, Sao Paulo, Brazil) attached to a digital camera model NewValue 48 Megapixel (Panasonic, Sao Paulo, Brazil). The calculation of the percentage of deaths adopted the Abbott formula, where:

$$\% M_x = (M_x - M_{C-} / 100 - M_{C-}) \, x \, 100 \quad (1)$$

Where $\% M_x$= corrected percentage of dead/immobile brine shrimp per test group or positive control; M_x= percentage of dead/immobile brine shrimp per test or positive control group; M_{C-}= percentage of dead brine shrimp in the negative control.

To estimate the representative value of the lethal concentration for 50% of the artemia (LC_{50}), the Probit method was applied, using the linear regression equation defined in Eq.2:

$$y = \beta_0 + \beta_1 . x \quad (2)$$

Where y = dependent variable or mortality, considered 50 for determining the LC_{50}; β_0 = intercept of the regression equation, calculated by $Y_{mean} - (\beta_1 \text{ x } X_{mean})$; β_1 = slope of the regression equation, calculated by $\sum (X - X_{mean}) (Y - Y_{mean}) / \sum (X - X_{mean})^2$; x = independent variable or concentration. R2 data were calculated by Jamovi software version 2.3.28 (Jamovi Project, Australia). The threshold for the presence of acute toxicity was adopted as a LC_{50} at concentrations <1,000 µg/mL.

Results

Saps of Croton lechleri present organoleptic differences

The initial organoleptic results showed that the burgundy color was the most prevalent in five samples, followed by dark brown in two samples and amber or colorless in one sample each. In only two samples, small

residues were detectable near the resins at the time of their dripping. A soapy effect was common, detectable in five samples. Viscosity analysis showed fluidity in seven samples, with only two being sticky. The most prevalent odor was woody, with only two exceptions being fruity or fragrant (Table 2).

Table 2. Organoleptic analysis of dragon's blood saps of *Croton lechleri*.

Criteria	*G1*	*G2*	*G3*	*G4*	*G5*	*G6*	*G7*	*G8*	*G9*
Color	Burgundy to red	Burgundy	Burgundy	Dark brown	Burgundy	Amber	Dark brown	Colorless	Burgundy
Residues	Yes	Yes	No	No	No	No	No	No	No
Soapy effect	Yes	No	No	Yes	Yes	No	Yes	No	Yes
Viscosity	Fluid	Sticky	Sticky	Fluid	Fluid	Fluid	Fluid	Fluid	Fluid
Odor	Woody	Woody	Woody	Woody	Woody	Woody	Fruity	Fragrant	Woody

Source: Research data.

Moderate spreading and density and intense dehydration

In microscopic analysis, the morphology at T0 showed droplets initially arranged in two phases in six samples (G1, G2, G4, G5, G6 and G9), with a larger central portion and a smaller peripheral halo that was generally less colored, which tended to coalesce into a single phase at T3. G8 was colorless but when low illumination was used against the stage, giving a grayish appearance, it was possible to observe the same previous two-phase behavior, with a lighter halo. Two samples (G3 and G7) remained in a single phase throughout the test. Furthermore, most of the samples presented a friable appearance at T3, with centrifugal cracks, more evident and irregular in G2 and more discrete and linear in G4, G5, G7 and G9. These findings could suggest hybrid two-phase samples with hydrophilic and hydrophobic charge to justify this visual separation compared to single-phase samples with more homogeneous compositions. The color also became darker in G1, G2, G4, G5, G7 and G9. All samples demonstrated some visual degree of superficial spreading, with saps from less spreading (G6) to greater spreading (G8). (Figure 1).

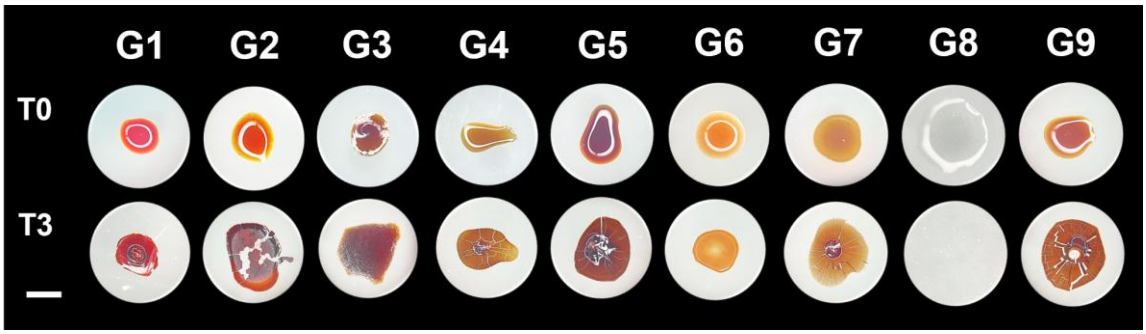


Figure 1. Dragon's blood saps of *Croton lechleri*, spreading at initial time (T0) and after 3h (T3). Scale bar=2mm. Source: Research data.

The spreading area of the saps followed a trend of moderate increase, with a relative difference greater than 48% in most samples. However, in G1 this value was well below that of all the others. For G8 and G9, the increase was more discrepant and exceeded twice the initial area. Only in G6 was there a slight decrease in the area. The phenomenon observed regarding mass was inversely proportional to the area, with the relative difference reducing by more than 59% in most groups, representing very evident dehydration, except in G3 and G8 with a reduction of up to 22%, which remained more liquid. The densities of the raw saps were more like each other and slightly higher than the typical density of water, reaching values equal to or greater than 1.2, despite divergences for G2 with a value above the standard and for G8 with a value below the others (Table 3).

Table 3. Analysis of spreading area, density and mass of dragon's blood saps of *Croton lechleri*.

Criteria	***G1***	***G2***	***G3***	***G4***	***G5***	***G6***	***G7***	***G8***	***G9***
Area at T0 (mm^2)	*3.421*	*4.747*	*4.193*	*3.753*	*5.766*	*5.858*	*5.699*	*10.570*	*5.278*
Area at T3 (mm^2)	*4.084*	*8.834*	*7.980*	*6.034*	*8.534*	*4.847*	*8.772*	*25.631*	*10.808*
Area difference (%)	*19.38*	*86.10*	*90.32*	*60.78*	*48.01*	*-17.26*	*53.92*	*142.49*	*104.77*
Mass at T0 (g)	*0.649*	*0.765*	*0.695*	*0.632*	*0.597*	*0.692*	*0.576*	*0.479*	*0.672*
Mass at T12 (g)	*0.146*	*0.145*	*0.538*	*0.138*	*0.152*	*0.119*	*0.231*	*0.404*	*0.097*

Source: Research data.

Alkaloids, polyphenols and terpenes are the most identifiable metabolites

Samples of G3 and G8 were excluded from Raman spectroscopy because they did not solidify, a condition that prevents measurement by the technique due to water interference in the analytes.

Raman spectroscopy analysis revealed similar vibrational band profiles among samples G1, G4, G5, G6, and G9, while sample G2 exhibited a slight shift of the bands to lower frequencies and reduced intensities. Sample G7 presented a lower signal-to-noise ratio, to mitigate this limitation, 25 accumulations per region were applied, followed by baseline correction and parabolic subtraction, although residual noise was not completely eliminated from the spectrum.

More prominent peaks were observed in G1, G2, G4, G5, G6 and G9 in the 1600–1500 cm^{-1} region, consistent with C=C aromatic ring stretching vibrations characteristic of alkaloids such as taspine and polyphenols such as tannins, as well as in the 1400–1300 cm^{-1} region, consistent with C=C vibrations associated with terpene structures.

Peaks above 1500 cm^{-1} were particularly relevant for the tentative characterization of aromatic compounds and polyphenols. In sample G5, the peak at 1556 cm^{-1} is consistent with aromatic structures, while the peak at 1574 cm^{-1} in sample G4 may suggest the presence of terpenic constituents. Peaks observed between 1455 and 1405 cm^{-1} across all samples are consistent with methyl group ($–CH_3$) vibrations associated with alkaloids such as taspine, while peaks around 1318–1320 cm^{-1} in G1, G5, and G9 may be attributable to carbon ring structures containing C–C and C–H bonds, as found in aromatic compounds and flavonoids such as catechins. In sample G2, bands in the 1250–1000 cm^{-1} region are consistent with C–N vibrations characteristic of alkaloids, including taspine. Overall, the vibrational signatures of the saps are collectively consistent with the presence of taspine, terpenes, catechins, tannins, and polyphenols, while no spectral features suggestive of saponin glycosides were identified in any of the samples. (Figure 2).

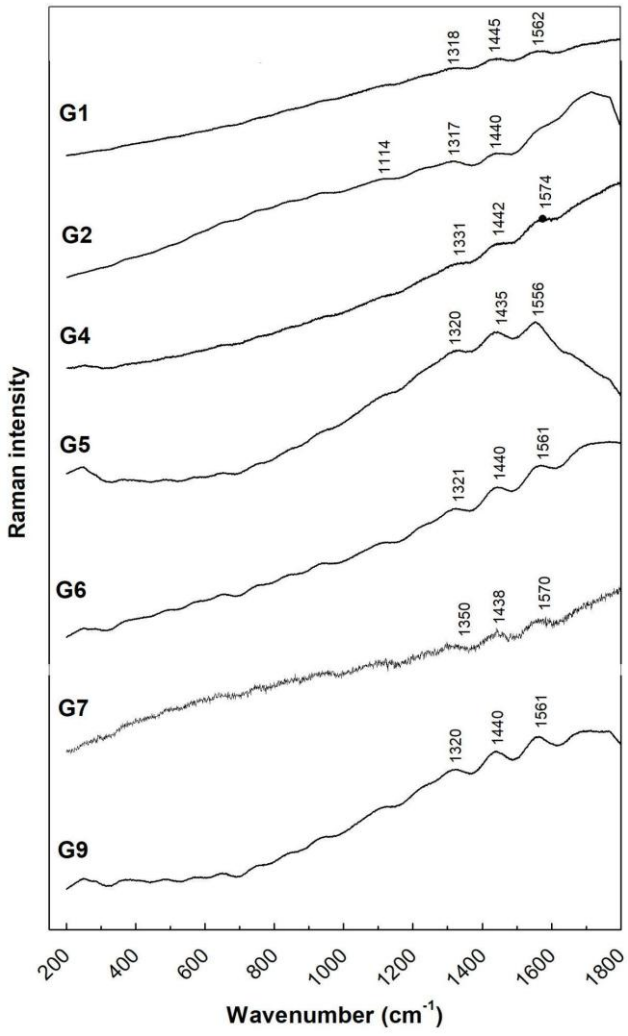


Figure 2. Raman spectrometry of dragon's blood saps of *Croton lechleri*. Source: Research data.

Phytochemical assays indicated convergent results regarding the absence of phenols, hydrolyzable tannins, anthocyanins, anthocyanidins, flavones, leucoanthocyaninides, triterpenoids, quinones and coumarins. The vast majority of samples (G1, G2, G4, G5, G6, G7 and G9) exhibited the presence of condensed tannins, chalcones, aurones, flavonols, flavanones, flavanonols, xanthones (by the sensitivity of the method with Mg), catechins (positive with 1% vanillin), steroids, diterpenes, saponins and alkaloids (positive with Dragendorff and Bouchardat). There were small variations in G3, with the absence of chalcones, aurones, steroids and diterpenes. The most discrepant sample was G8, with only the presence of free steroids and the absence of all other secondary metabolites (Table 4).

Table 4. Phytochemical prospecting of commercial samples of dragon's blood saps of *Croton lechleri*.

Secondary metabolite	Reagent	G1	G2	G3	G4	G5	G6	G7	G8	G9
Phenols	$FeCl_3$	-	-	-	-	-	-	-	-	-
Hydrolyzable tannins	$FeCl_3$	-	-	-	-	-	-	-	-	-
Condensed tannins	$FeCl_3$	+	+	+	+	+	+	+	-	+
Anthocyanins	HCl and NaOH	-	-	-	-	-	-	-	-	-
Anthocyanidins	HCl and NaOH	-	-	-	-	-	-	-	-	-
Flavones	HCl and NaOH	-	-	-	-	-	-	-	-	-
Flavonols	HCl and NaOH	-	-	-	-	-	-	-	-	-
Xanthones	HCl and NaOH	-	-	-	-	-	-	-	-	-
Chalcones	HCl and NaOH	+	+	-	+	+	+	+	-	+
Aurones	HCl and NaOH	+	+	-	+	+	+	+	-	+
Flavononols	HCl and NaOH	-	-	-	-	-	-	-	-	-
Leucoanthocyanidins	HCl and NaOH	-	-	-	-	-	-	-	-	-
Catechins	HCl and NaOH	-	-	-	-	-	-	-	-	-
Flavanones	HCl and NaOH	+	+	-	+	+	+	+	-	+
Flavonols, Flavanones, Flavanonols and Xanthones	Mg and HCl	+	+	+	+	+	+	+	-	+
Catechins	1% Vanillin and HCl	+	+	+	+	+	+	+	-	+
Free steroids	H_2SO_4, $(CH_3CO)_2O$ and Na_2SO_4	+	+	-	+	+	+	+	+	+
Triterpenoids	H_2SO_4, $(CH_3CO)_2O$ and Na_2SO_4	-	-	-	-	-	-	-	-	-
Diterpenes	10% $Cu(CH_3COO)_2$	+	+	-	+	+	+	+	-	+
Saponins	Shaking	+	+	+	+	+	+	+	-	+
Quinones	10% NaOH	-	-	-	-	-	-	-	-	-
Alkaloids	Dragendorff	+	+	+	+	+	+	+	-	+
Alkaloids	Bouchardat	+	+	+	+	+	+	+	-	+
Coumarins	10% NaOH	-	-	-	-	-	-	-	-	-

Positive (+) or negative (-) results. Source: Research data.

Great toxicological safety in tests with microcrustaceans

The LC_{50} was well above 1000 μg/mL in each of the experimental conditions, confirming the absence of acute toxicity of the saps. There were intergroup differences in mortality caused by exposure of *Artemia salina* to the different dragon's blood saps, with discrepancies for G6 with a lower LC_{50} and for G5 and G8 with higher LC_{50}. However, there were differences between all samples for potassium dichromate, which showed

high mortality at a minimum concentration. Linear regression of the data followed an increasing linear pattern whose positive slope showed that the slight increase in the percentage of Artemia deaths was directly proportional to the concentration of the sap under test. The higher values of the coefficients of determination (R2) for the saps ranging from 52% to 93% more robustly explain the increase in concentration as a predictor of mortality, with the model presenting a better fit to the data. For potassium dichromate, the regression model explains little of the variation around the mean in the response data. (Table 5)

Table 5. Acute toxicity to *Artemia salina* calculated by mortality, linear regression and lethal concentrations (LC_{50}) of dragon's blood saps of *Croton lechleri* (G1–G9) and potassium dichromate (PD, control).

Material	Average mortality (%)					Linear regression (R^2)	Linear regression (Equation)	LC_{50} (µg/mL)
	10 µg/mL	100 µg/mL	500 µg/mL	1,000 µg/mL	5,000 µg/mL			
G1	0.00± 0.00	0.00± 0.00	1.33± 0.02	8.05± 0.05	23.38± 0.05	0.81	Y=0.343848378+ 0.004696333X	10,573.39
G2	0.00± 0.00	0.00± 0.00	0.00± 0.00	12.43± 0.04	20.05± 0.07	0.63	Y=1.23299898+ 0.003980787X	12,250.59
G3	0.00± 0.00	1.78± 0.02	2.91± 0.03	8.34± 0.03	16.61± 0.05	0.67	Y=1.864407728+ 0.003073822X	15,659.85
G4	0.00± 0.00	2.18± 0.03	4.16± 0.05	15.29± 0.09	23.18± 0.07	0.52	Y=3.358994286+ 0.00423828X	11,004.70
G5	0.00± 0.00	0.98± 0.02	2.46± 0.02	4.38± 0.05	11.74± 0.02	0.57	Y=1.073077677+ 0.002145024X	22,809.49
G6	0.00± 0.00	1.05± 0.02	2.13± 0.03	17.11± 0.04	48.25± 0.05	0.93	Y=0.96193381+ 0.009641502X	5,086.14
G7	0.00± 0.00	1.29± 0.02	1.85± 0.02	5.95± 0.07	24.15± 0.06	0.75	Y=0.323055155+ 0.004783166X	10,385.79
G8	0.00± 0.00	0.00± 0.00	0.91± 0.01	1.54± 0.02	7.94± 0.04	0.57	Y=-0.032108243+ 0.001594938X	31,369.31
G9	0.00± 0.00	0.00± 0.00	0.00± 0.00	3.53± 0.03	14.38± 0.05	0.75	Y=-0.32286915+ 0.002952851X	17,042.13
PD	6.42± 1.37	52.67± 9.39	76.12± 11.99	93.82± 0.64	100.00± 0.00	0.38	Y= 50.54808223+ 0.011541661X	47.49

Source: Research data.

Discussion

The organoleptic description of dragon's blood generally prioritizes the visual aspect of the sap, which is characteristically reddish (Edwards et al. 2004; Tezza 2025; Gupta et al. 2008; Albuquerque et al. 2025) and has a soapy effect (Gupta et al. 2008; Albuquerque et al. 2025; Timilsena et al. 2023). The phenomenon of microscopic darkening of most dehydrated saps could be justified by contact with oxygen, although there is no scientific evidence to support this hypothesis. Other non-Croton dragon's blood resins have demonstrated a composition rich in aromatic rings (Edward et al. 2001), but this study contributes with an unprecedented olfactory comparison between the different Croton saps, with a woody tone compatible with their plant origin. The presence of small residues in the storage bottle in some samples highlights the importance of quality control of *Croton lechleri* latex (Albuquerque et al. 2025).

Wettability is the ability of a liquid to maintain contact with a solid surface, which may exhibit hydrophobicity, hydrophilicity, oleophilicity or oleophobicity (Sun et al. 2021). On nonpolar plastic surfaces of the stereomicroscope, Croton saps presented more spread-out droplets, suggesting greater hydrophobic or oleophilic content, while the few that presented more concentrated droplets with little dispersion suggest greater hydrophilic or oleophobic content. Based on these properties, most saps, particularly G2, G5 and G7, may be better suited for cutaneous applications as emollients, to retain moisture and replace lost natural oiliness given their fatty acid composition typical of vegetable oils, or in association with creams as occlusives, as a final layer of thicker protection (Gil-Castaño & Cardona 2020). Among the few hydrophilic samples, their use in combination with biomaterials such as hydrogels for wound lining and healing (Wang et al. 2023) or collagen matrices stands out—reaching maximum swelling after 10 to 30 minutes of contact with simulated biological fluids or exudates. The hydrophilicity of scaffolds for intraosseous implantation in orthopedic and dental surgeries has demonstrated positive modulation for the adsorption of proteins, immune cells, angiogenesis and osteogenesis, favoring successful integration into the tissue bed (Wei et al. 2024). However, it is worth noting that the internal body effects of *Croton lechleri* involving pharmacokinetics and pharmacodynamics are not fully known, which directs its use with parsimony, given the scarcity of clinical scientific evidence of the safety and efficacy of this dragon's blood (Gupta et al. 2008; Albuquerque et al. 2025; Peres et al. 2023).

The phenolic or antioxidant components of *Croton lechleri* sap can be degraded by up to 20% by continuous exposure to ultraviolet radiation and also change their properties under relative humidity, being more stable at room temperature of 21°C than refrigerated at 4°C for up to 39 days without compromising their effectiveness (Escobar et al. 2018). Based on the conservation of the samples in translucent or opaque bottles and at room temperature for the tests, the differences in color and shape observed in some groups over hours of contact between sap and air could be explained both by the antioxidant reaction of the constituents of each sap and by some stage of its packaging and storage that influenced the quality of the natural product. Although commercial samples give an average shelf life of between 2 and 3 years, a study suggests that the storage life of *Croton lechleri* latex is 3 to 6 months to preserve its properties in medicinal applications (Peres et al. 2023), which raises the question of the correct shelf life for this type of natural product, although there is still no consensus on the matter.

The secondary metabolites found in the dragon's blood saps tested generally converge with the literature and from them some of their biomedical applicability can be interpreted. The presence of diterpenoids is quite common (Silva et al. 2022; Hidalgo et al. 2018; Rodríguez-Garcia et al. 2010; Monterubbianesi et al. 2023; Albuquerque et al. 2025), since they constitute up to 85% of the compounds found in species of the genus Croton, with a broad spectrum of bioactivities, including cytotoxic, anti-inflammatory, antifungal, acetylcholinesterase inhibitory, neurite growth promoting (Xu et al. 2018) and antitumor properties (Monteiro et al. 2005). Studies indicate that up to 90% of the dry weight of *Croton lechleri* latex corresponds to polyphenols, including diterpenes, catechins and proanthocyanidins, with antioxidant action (Lopes et al. 2013; Albuquerque et al. 2025).

The presence of condensed tannins, which are more stable plant polyphenols than hydrolysable ones, would promote favorable anti-inflammatory, antimicrobial and healing effects, and their typically reddish pigments could justify the hue of dragon's blood sap (Albuquerque et al. 2025; Monteiro et al. 2005). The presence of flavonoids and chalcones is coherent, as they are also the main components of *Croton lechleri* sap with anti-inflammatory and antioxidant effects (Hidalgo et al. 2018; Gupta et al. 2008; Peres et al. 2023; Escobar et al. 2018). Saponins are plant products with detergent and emulsifying effects, related to anti-inflammatory and antimicrobial actions, and foam production is a simple control of their presence and the quality of the sap (Gupta et al. 2008; Albuquerque et al. 2025; Timilsena et al. 2023). The evidence of alkaloids reinforces the dark hue and antioxidant activity of dragon's blood, especially attributed to taspine (Albuquerque et al. 2025; Escobar et al. 2018; Xu et al. 2018). Taspine is cited as the only alkaloid found in the latex of adult *Croton lechleri* trees, although other types of alkaloids exist in its leaves and bark (Lopes et al. 2013; Milanowski et al. 2002) and constitutes an active principle with antimicrobial and healing action, which further reinforces its importance (Londoño-Lemos & Bustamante 2019; Martins et al. 2016; Hidalgo et al. 2018; Cayo & Barrera 2014; Monterubbianesi et al. 2023; Gupta et al. 2008). Phytosteroids can be found in dragon's blood saps, with cellular anabolic action and immunomodulatory potential (Rodríguez-Garcia et al. 2010; Albuquerque et al. 2025; Salatino et al. 2007), which combines the samples, all positive for the metabolite, with this mechanism of action.

On the other hand, the initial negative tests for phenols, anthocyanins and anthocyanidins could hypothesize a lower antioxidant action, since these are compounds expected in dragon's blood sap (Londoño-Lemos & Bustamante 2019; Hidalgo et al. 2018; Rodríguez-Garcia et al. 2010; Monterubbianesi et al. 2023; Albuquerque et al. 2025; Peres et al. 2023). Triterpenoids can be found in *Croton lecheri* sap and exhibit antifungal action (Lopes et al. 2013; Peres et al. 2023; Xu et al. 2018; Salatino et al. 2007), but their presence was negative in all samples tested, which refutes the literature. Quinones are oxidant and electrophilic compounds that generate antimicrobial action (Sousa et al. 2016), which can be found in *Croton lechleri*, although it is not a typical finding (Lopes et al. 2013; Peres et al. 2023). Coumarin is a very versatile natural product, with anticoagulant, anticancer, antioxidant, antiviral, antidiabetic, anti-inflammatory, antimicrobial and anti-neurodegenerative agent action (Franco et al. 2021), but this metabolite is not usually found in *Croton lechleri* sap (Xu et al. 2018; Salatino et al. 2007). Despite the wide range of uses, there is a mixture of several phytochemical compounds in the raw extracts, which requires that the interpretation of the general potential of the Amazonian sap be done with parsimony and in safe doses (Silva et al. 2022; Gupta et al. 2008).

Raman spectroscopy can suggest a range of plant products, such as carotenoid pigments (Udensi et al. 2022; Allakhverdiev et al. 2022), lipids (Allakhverdiev et al. 2022), minerals, and secondary metabolites (Allakhverdiev et al. 2022; Saletnik et al. 2021). In the latter case, the most expressive range is 1110-1600 cm^{-1}, with bands reaching 1000-3000 cm^{-1} (Saletnik et al. 2021). This suggests the presence of phenolic components — such as gallic acid and catechins, as found in white and red wines— in the 50-850 cm^{1} range, although it can extend to the range of 1600-1750 cm^{-1} (Deneva et al. 2019). In the species *Croton anisodontus Mull. Arg.*, the

natural substance 2-hydroxy-3,4,6-trimethoxyacetophenone, a precursor of chalcones, with antibacterial, anxiolytic, cytotoxic and antifungal properties, has a wide range of vibrational modes from 30-3100 cm^{-1}, which sometimes makes the identification of plant metabolites very challenging (Oliveira et al. 2014). The Raman spectra of *Croton lechleri* from this study are close to the expected results for Croton sp., *C. gossipifolius*, and *C. aromaticus*, with a more pronounced vibrational signature, suggesting bands at 1672 cm^{-1} for C=O and C=C, at 1612 cm^{-1} with vibration for C=C and C=O, at 1338 cm^{-1} with weak to medium and broad vibration for C–C and CH_2, at 1292 cm^{-1} with weak to medium or broad vibration for CH_2, and at 784 cm^{-1} with vibration for CCH (Edwards et al. 2004). Raman spectra indicating alkaloids, terpenes, and polyphenols in the analyzed *Croton lechleri* sap converge with the approximate wavenumber, vibrational intensity, and correlation of the chemical groups present in the constitution of plant secondary metabolites (University of California, 2025).

The Raman spectra of other trees also called dragon's blood scattered throughout the world, *Eucalyptus terminalis* from Australia, *Dracaena cinnabari* from Socotra in East Africa and *Daemonorops draco* from the Caribbean exhibited bands at 3100-2800 cm^{-1} followed by 1750-700 cm^{-1}, with the identification of benzoic acid being more evident at 1000, 800 and 1600 cm^{-1}, an intermediate in the biosynthesis of many secondary metabolites, but with a different vibrational signature from the genus Croton (Edward et al. 2001). Raman spectrometry of ancient or archaeological resins makes it difficult to build a database for identification by conservation status, but recent non-Croton dragon's blood resins had spectral signatures that allowed their discrimination by the technique (Edwards & Ali 2011). In this way, the variation of less defined bands and spectra among the tested groups of *Croton lechleri* could represent a reflection of age or degradation of the resin, due to thermal or environmental effects. These thermal effects are directly related to decomposition during analysis; due to the intense red color of the resins, they absorb a significant amount of energy, which causes the use of a laser to cause immediate decomposition of the sample (Edwards et al. 2004).

The acute non-toxicity of dragon's blood sap is based on the classic test by Meyer et al. (1982), which found that more than 50% of brine shrimp died under highly toxic conditions at low concentrations of plant extracts, such as $LC50 \leq 30$ μg/mL, with decreasing acute toxicity at higher concentrations, with an extract considered non-toxic from $LC50 \geq 1000$ μg/mL. This is the first report in the literature on the acute non-toxicity effect of *Croton lechleri* in the *Artemia salina* model, although other species of the genus Croton exhibit more evident toxicities.

The essential oil of leaves of the Amazonian plant *Croton campinarensis* from the northern region of Brazil exhibited an LC_{50} of 20.84 μg/mL (Costa et al. 2022), while the essential oil of leaves of *Croton rudolphianus*, a plant typical of the caatinga or northeastern region of Brazil, exhibited an LC_{50} of 68.33 μg/mL (Ribeiro et al. 2021). Similarly, the leaf extract of *Croton blanchetianus*, also typical of the caatinga biome, exhibited an LC_{50} of 66.26 μg/mL, with 2,5-dihydroxybenzoic acid in high concentrations being attributed as the main component responsible for this acute toxicity (Figueiredo et al. 2022). Methanolic extracts of bark of *Croton rhamnifolioides* from the Brazilian caatinga exhibited an LC_{50} of 486.59 μg/mL (Lima et al. 2015). The ethanolic extract of the bark of *Croton heliotropiifolius*, a species typical of the caatinga, exhibited moderate toxicity, with an LC_{50} of 396.6 μg/mL (Silva et al. 2020), while its methanolic extract of leaves showed an LC_{50} of 637.29 μg/mL (Silva et al. 2018). *Croton floribundus*, a plant typical of the Atlantic Forest biome located in the Southeast and South regions of Brazil, also showed differences in acute toxicity depending on the extraction site, with ethanolic extracts of the stem bark exhibiting high acute toxicity, with an LC_{50} of 89.6 μg/mL, while the leaf extract did not exhibit acute toxicity, with an LC_{50}>1000 μg/mL (Barth et al. 2018). The saps of *Croton lechleri* were shown to be safe and non-toxic, with an LC_{50} well above that found in the bark of trees of the genus Croton with moderate toxicity. In other biological systems, J774 murine macrophages exposed to *Croton lechleri* exhibit low LC_{50} of 25 μg/mL, which may link the role of dragon's blood in both controlling the inflammatory and repair processes (Silva et al. 2022). In human cells, acute toxicity to ethanolic extracts of *Croton lechleri* suggests an association with higher LC_{50} in normal human cells including the renal cell line VERO (218.92 μg/mL) and epidermal HaCaT (205 μg/mL), being lower in tumors in HeLa uterine (17μg/mL), MDA-MB231 breast, HepG2 liver and SW-480 colon cancer cell lines (>50μg/mL) (Silva et al. 2022). The dose for human internal use of *Croton lechleri* has not yet been fully established, but through an allometric scale, a conversion from animal models to humans is proposed, reaching an equivalent oral dose of 32.43 mg/kg, with no associated acute toxicity and with anti-inflammatory efficacy (Tezza 2025).

Conclusion

The saps of *C. lechleri* investigated in this study exhibited variability in organoleptic characteristics, while showing similar tendencies in dispersion, density, and dehydration behavior. Phytochemical screening indicated

a predominance of alkaloids, polyphenols, and terpenes as the main classes of secondary metabolites across the samples. The *Artemia salina* bioassay suggested absence of acute toxicity, providing a preliminary indication of toxicological safety, though further preclinical studies are needed to confirm these findings. The characterization data obtained here support the relevance of continued investigation into the mechanisms of action and dose standardization of dragon's blood saps, as necessary steps toward validating their safe and effective use in future preclinical and clinical applications.

Acknowledgements

The authors thank the Biomaterials Laboratory (BIOMAT, Postgraduate Program in Biotechnology, Federal University of Ceará), the Center for Bioactive Phytochemical Studies (NEFB, State University of the Valley of the Acarau), the Multidisciplinary Laboratory in Dental Research (LAMPO, Postgraduate Program in Dentistry, Federal University of Ceará) and the technician Manoel Florindo Júnior for making the experimental analyses possible.

Funding

This research received external funding from Cearense Foundation for Scientific and Technological Development Support (FUNCAP-Brazil, research grant BPI 04/2022, process n. BP5-0197-00060.01.00/22), Coordination of Improvement of Higher Education Personnel (CAPES-Brazil, doctoral scholarship, code 001), and National Council for Scientific and Technological Development (CNPq-Brazil, scientific initiation scholarship, code 138704/2024-4).

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