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Phytochemical prospection and antibacterial activity of native plants from the cerrado of goiás, Brazil

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Abstract
The Brazilian biome Cerrado is a big source of obtaining plants for the research of new bioactive compounds with antibacterial potential. In this study, methanolic extracts of the leaves from *Stryphnodendron adstringens* (Mart) Coville and *Solanum lycocarpum* A. St-Hil; and ethanolic extracts of *Anadenanthera falcata* (Benth.) Speg. Stem and *Cochlospermum regium* (Mart. ex Schrank) Pilg. Tuberous roots were submitted to phytochemical prospecting and determination of antibacterial activity against four bacterial cultures *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* by the Kirby-Bauer method of plate diffusion. Diverse groups of secondary metabolites and some constituents were identified, suggesting bioavailability of alkaloids, anthraquinones, catechins, coumarins, steroids and triterpenoids, phenols and tannins, flavonoids, anthocyanins and anthocyanidins, flavonols, flavonones, flavanonols and xanthones, quinones and saponins. Likewise, all extracts presented antibacterial potential, especially *Stryphnodendron adstringens* (Mart) Coville extract at the concentration of 10 mg/mL against *Escherichia coli* and *Staphylococcus aureus*.

Keywords: Phytochemical prospection, antimicrobial activity, crude extracts, Brazilian biome Cerrado

1. Introduction
The use of medicinal plants has socioeconomic relevance on the quality of life of low-income communities owing to their availability, low toxicity and minimal risk of side effects. This is a common reality in rural Brazil, also associated with difficulties in accessing basic public health services (Bessa *et al.*, 2013) [1].

In view of the great Brazilian biodiversity, the Cerrado biome is one of the main sources of obtaining plants for the research of new bioactive compounds with antibacterial potential. However, research is still scarce and with future availability compromised due to threats such as deforestation and fires, even reaching Legal Reserve Areas, which has led to the loss of medicinal biodiversity (Silva; Miranda; Conceição, 2010) [2]. Many of these plants used in traditional medicine are possible sources of substances with antimicrobial activities against microorganisms harmful to human health, such as *Anadenanthera falcata* (Benth.) Speg. (*A. falcata*), *Cochlospermum regium* (Mart. ex Schrank) Pilg. (*C. regium*), *Stryphnodendron adstringens* (Mart) Coville (*S. adstringens*), and *Solanum lycocarpum* A. St-Hil (*S. lycocarpum*) (Fig 1).

A. falcata (popularly known as angico-do-cerrado) is a specie with wide geographical distribution throughout the Brazilian Cerrado biome. It belongs to the Leguminosae family and the subfamily Mimosoideae (Carvalho, 2003) [3]. The plant bark is used in the form of teas by folk medicine to combat cough and bronchitis, as a healing, antibacterial, in the treatment of wounds and ulcerations, angina and dysentery (Lorenzi, 2002) [4].

C. regium (popularly known as algodãozinho-do-cerrado) is a native and non-endemic plant, belonging to Bixaceae family, subfamily Cochlospermaceae and it is distributed along the extension of the Brazilian territory. Sprout in open areas, mainly in the Cerrado biome and vacant lots (Ribeiro *et al.*, 2017) [5]. In folk medicine, *C. regium* tuberous roots are used as treatment for gastritis and stomach ulcers, salpingitis, endometritis, cervicitis, arthritis, dysmenorrhea, as carminative, and anti-acne (Arunachalam *et al.*, 2019) [6].

S. adstringens (popularly known as barbatimão) is a native species belonging to Fabaceae family and the subfamily Mimosoideae (Panizza *et al.*, 1988) [7]. It is widely distributed in the Brazilian Cerrado biome and is used by folk medicine as an antibacterial, anti-inflammatory,

antiseptic, astringent and healing agent (Luiz *et al.*, 2015) [8]. *S. lycocarpum* (popularly known as lobeira) belongs to the Solanaceae family and is a plant also widely distributed by Brazilian Cerrado biome, especially in the central-west region (Lorenzi, 2002) [4]. In folk medicine, *S. lycocarpum* is used as a sedative, antiasthmatic, antidiabetic, anti-epileptic, dyslipidemic, hypocholesterolemic and in the treatment of abdominal pain and renal colic (Munari *et al.*, 2012) [9].

Fig 1: Botanical structures of *A. falcata*: plant (A), stem (B); *C. regium*: plant (C), leaves and tuberous roots (D); *S. adstringens*: plant (E), leaves (F); *S. lycocarpum*: plant (G), leaves and fruit (H).
Source: Authors, 2019.

Given the context, this study aimed to perform phytochemical prospections and verify possible antibacterial activities of four native plants from the Cerrado biome of Goiás, Brazil.

2. Material and Methods

Location and characterization of the study area

The mesoregion in the center of Goiás (Fig 2) is one of the five meso regions of the state of Goiás. It has an area of

40,836.704 km² and is formed by the union of 82 municipalities grouped in eighteen microregions. The microregion 09 (Fig 3) composed of Adelândia, Americano do Brasil, Anicuns, Aurilândia, Avelinópolis, Buriti de Goiás, Firminópolis, Mossâmedes, Nazário, Sanclerlândia, Santa Bárbara de Goiás, São Luís de Montes Belos and Turvânia was the area of collection and study of the plants in question.

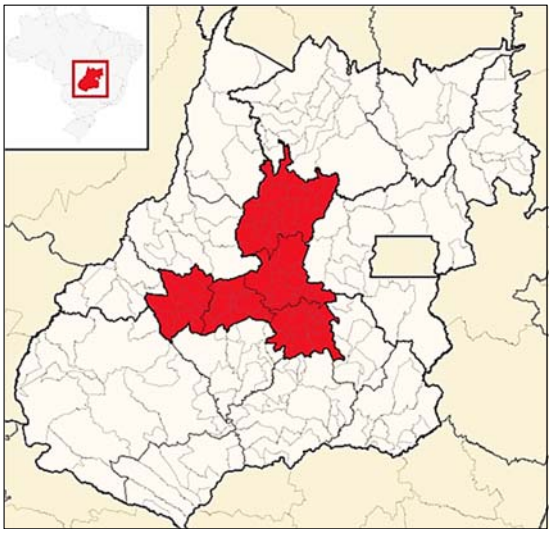

Fig 2: Geographic location of the mesoregion in the center of Goiás, state of Goiás, Brazil, 2019.
Source: IBGE, 2019 [10].

Fig 3: Geographic location of microregion 09, mesoregion of central Goiás, state of Goiás, Brazil, 2019.
Source: IBGE, 2019 [10].

Botanical species

A. falcata (angico-do-cerrado), *C. regium* (algodão-do-cerrado), *S. lycocarpum* (lobeira) and *S. adstringens* (barbatimão), native species of the Cerrado biome were selected for this study due to the popular uses and indications for possible antimicrobial effects. Part of the stem of a *A. falcata* (Latitude: 16.21029°S; Longitude: 50.32181°W), tuberous roots of *C. regium* (Latitude: 16.53460°S; Longitude: 50.35924°W), and leaves of *S. adstringens* (Latitude: 16.17469°S; Longitude: 50.30809°W) and *S. lycocarpum* (Latitude: 16.16051°S; Longitude: 50.26272°W) were collected in March, 2019. Exsiccates from each plant were identified by Prof. Dr. Edvande Xavier dos Santos Filho and deposited in the herbarium of the University Center Montes Belos (School Farm).

Plant extracts obtainment and preparation

400 g of stem from *A. falcata*, 400 g of tuberous roots from *C. regium*, 400 g of leaves from *S. adstringens* and 400 g of leaves from *S. lycocarpum* were collected, taken to the Laboratory of Pharmacognosy in the University Center Montes Belos, weighed and submitted to kiln-drying (Odontobras EL 1.3) at 40±0.5 °C for 3 days. Subsequently, vegetable samples were ground in a Wiley knife micro mill (Tecnal TE-648) and extracts were prepared from 100 g of the ground powder of each. Next, *A. falcata* and *C. regium* were extracted by maceration in 500 mL of 95% ethanolic solution; and *S. adstringens* and *S. lycocarpum* in 500 mL of 95% methanolic solution, all for 7 consecutive days. The crude extracts were subsequently concentrated on a rotary evaporator under reduced pressure (up to 50 °C) and weighed. Then, extracts' concentrates were dried in a hot-air oven at the temperature of 50±0.5 °C for 24 hours and weighed again to calculate the yield (%) of the concentrated crude extracts and after drying. The ratio between the mass (g) of the concentrated crude extract (w) and after drying (w) was applied (Bessa *et al.*, 2013) [1].

Phytochemical prospecting of plants constituents

Crude ethanolic extracts of *A. falcata* and *C. regium*; and crude methanolic extracts of *S. adstringens* and *S. lycocarpum* were subjected to different analyzes for phytochemical characterization according to (Bessa *et al.*, 2013) [1] and (Matos, 1997) [11] with adaptations. The positivity for each group of secondary metabolites and constituents analyzed was achieved from the development of color and/or precipitate, characteristic for analysis of each class.

Secondary Metabolite Groups Analyzed

- Alkaloids;
- Anthraquinones;
- Catechins;
- Coumarins;
- Steroids and triterpenoids;
- Phenols and Tannins;
- Flavonoids, Anthocyanins, Anthocyanidins;
- Flavonols, flavanones, flavanonols and xanthones;
- Quinones;
- Saponins;

Determination of antibacterial activity

Bacterial culture and standardization of concentrations of the crude extracts

Initially, to determine the antibacterial activity of crude ethanolic extracts of *A. falcata* and *C. regium*; and crude methanolic extracts of *S. adstringens* and *S. lycocarpum*, the Kirby-Bauer method of plate diffusion was applied (Gonçalves *et al.*, 2016) [12]. Four bacterial cultures were used, one Gram-positive: *Staphylococcus aureus* (ATCC 25923); and three Gram-negative: *Escherichia coli* (ATCC 25922), *Klebsiella pneumoniae* (ATCC 13883) and *Pseudomonas aeruginosa* (ATCC 27853). As a negative control, the solvent ethanol (equivalent to the highest concentration of the extract used) was applied; and as a positive control, the antibacterial norfloxacin 400 mg was used. To determine the sensitivity of bacterial strains to the concentrations of crude extracts, serial

dilutions were performed: 20 mg/mL, 10 mg/mL, 5 mg/mL, 2,5 mg/mL, 1,25 mg/mL, 0,625 mg/mL, 0,312 mg/mL.

Antibacterial susceptibility test

Before proceeding to the susceptibility test, bacterial strains were activated in Brain Heart Infusion (BHI) broth medium at 35±2 °C for 24 hours. Next, an inoculum of each bacterial suspension was prepared in sterile saline based on the McFarland nephelometric scale (1 x 10^8 CFU.mL-1). Soon after, plates were prepared with the inoculated agar following the recommended by NCCLS - eighth edition [13], and filter paper discs (0.6 mm in diameter) previously sterilized with the pre-established concentrations of crude ethanolic extracts of *A. falcata* and *C. regium*; and crude methanolic extracts of *S. adstringens* and *S. lycocarpum* were impregnated on the agar surface. Then, the plates were incubated at 4 °C for 15 minutes to guarantee the diffusion of the extracts on agar, and finally incubated in an oven at 35 ± 2°C for 24 hours for subsequent observation of the inhibition halos. Analyzes were performed in triplicate.

Statistical Analysis

Results were expressed as mean ± standard deviation. Windows version of the Graph Pad Prism 5.01 software was used to perform statistical tests. And, one- or two-way ANOVA grouped analysis was used, followed by Bonferroni post-tests, with *P* values <0.05.

3. Results

Table 1 describes the botanical identification, popular name and methodological characteristics for preparation of crude extracts from each plant species.

Table 1: Methodological description for preparation of crude extracts from each plant species.

Characteristics				
Family	**Leguminosae**	**Bixaceae**	**Fabaceae**	**Solanaceae**
Specie	*Anadenanthera falcata* (Benth.) Speg.	*Cochlospermum regium* (Mart. ex Schrank) Pilg.	*Stryphnodendron adstringens* (Mart) Coville	*Solanum lycocarpum* A. St-Hil
Local name	Angico-do-cerrado	Algodãozinho-do-cerrado	Barbatimão	Lobeira
Vegetable part	Stem	Tuberous roots	Leaves	Leaves
Extraction method	Maceration	Maceration	Maceration	Maceration
Mass of vegetable material (g)	100g	100g	100g	100g
Solvent	Ethanolic solution 95%	Ethanolic solution 95%	Methanolic solution 95%	Methanolic solution 95%
Volume of solvent (mL)	500 mL	500 mL	500 mL	500 mL
Extraction time	7 consecutive days	7 consecutive days	7 consecutive days	7 consecutive days
Temperature	25±2 °C	25±2 °C	25±2 °C	25±2 °C

The sample of the crude ethanolic extract from the stem of *A. falcata* showed a yield (w/w) of 17.2%, with a determination of the extractives content of 31.1%. From the phytochemical prospecting, the presence of diverse groups of secondary metabolites and some constituents were identified, suggesting bioavailability of anthraquinones, catechins, steroids and triterpenoids, phenols and tannins, flavonoids, anthocyanins and anthocyanidins, flavonols, flavonones, flavanonols and xanthones, e quinones. Only coumarins and saponins were not detected (Table 2).

The specimen of the crude ethanolic extract from tuberous roots of *C. regium* demonstrated a yield (w/w) of 23.4%, with a determination of the extractives content of 6.4%. Phytochemical prospecting identified alkaloids, catechins, coumarins, phenols and tannins, flavonols, flavonones, flavanonols and xanthones, and quinones. Anthraquinones, steroids and triterpenoids, and saponins were not detected by tests (Table 2).

The specimen of the crude methanolic extract from leaves of *S. adstringens* demonstrated a yield (w/w) of 21.85%, with a determination of the extractives content of 12.3%. Phytochemical prospecting identified the presence of alkaloids, anthraquinones, catechins, steroids and triterpenoids, phenols and tannins, flavonoids, anthocyanins and anthocyanidins, flavonols, flavonones, flavanonols and xanthones, quinones and saponins. Only coumarins were not detected by the tests (Table 2).

And, the sample of the crude methanolic extract from leaves of *S. lycocarpum* showed a yield (w/w) of 23.62%, with a determination of the extractives content of 5.5%. From the phytochemical prospecting, steroids and triterpenoids, flavonoids, anthocyanins and anthocyanidins, flavonols, flavonones, flavanonols and xanthones were identified. Alkaloids, anthraquinones, catechins, coumarins, phenols and tannins, quinones and saponins were not detected (Table 2).

Table 2: Classes of secondary metabolites identified in the crude extracts of *A. falcata*, *C. regium*, *S. adstringens* and *S. lycocarpum* according to vegetable parts.

Classes of metabolites	***A. falcata* (stem)**	***C. regium* (root)**	***S. lycocarpum* (leaf)**	***S. adstringens* (leaf)**
Alkaloids	-	+	-	+
Anthraquinones	+	-	-	+
Catechins	+	+	-	+
Coumarins	-	+	-	-
Steroids and triterpenoids	+	-	+	+
Phenols and tannins	+	+	-	+
Flavonoids, anthocyanins, anthocyanidins	+	+	+	+
Flavonols, flavonones, flavanonols, xanthones	+	+	+	+
Quinones	+	+	-	+
Saponins	-	-	-	+
Determination of the extractives content (%)	31.1	6.40	5.5	12.3
Yield (w/ w)	68.9g	93.6g	94.5g	87.4g
(+, present); (-, absent)				

Results obtained in the study of antibacterial activity were satisfactory for the strains *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* from the crude ethanolic extract of *A. falcata*. Inhibition halos ranged from 1.27 ± 0.3 mm to 2.17 ± 0.4 mm. The most expressive result was against the gram-negative bacterial strain *Klebsiella pneumoniae* at the concentration of 2.5 mg/mL. *A. falcata* extract did not show antibacterial activity against *Escherichia coli* and *Staphylococcus aureus* at any tested concentrations (20-0.312 mg/mL).

Crude ethanolic extract from tuberous roots of *C. regium* demonstrated antibacterial activity for the strains *Escherichia coli* and *Staphylococcus aureus* with inhibition halos ranging from 1.22 ± 0.2 mm to 2.22 ± 1.1 mm. The most expressive result was against the gram-negative bacterial strain *Escherichia coli* at the concentration of 20 mg/mL. No antibacterial activity was observed against the strains *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* at tested concentrations (20-0.312 mg/mL).

Crude methanolic extract from leaves of *S. adstringens* inhibited the growth of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*. Inhibition halos ranged from 0.8 ± 0.1 mm to 10.53 ± 1.2 mm. The most expressive result was against the gram-negative bacterial strain *Escherichia coli* at the concentration of 10 mg/mL; and the least expressive, but still inhibiting growth, was related to *Pseudomonas aeruginosa* at the concentration of 5 mg/mL.

Lastly, crude methanolic extract from leaves of *S. lycocarpum* demonstrated activity against *Escherichia coli*, *Klebsiella pneumoniae* and *Staphylococcus aureus* with inhibition halos ranging from 1.14 ± 0.8 mm to 2.72 ± 1.01 mm. The most expressive result was against the gram-positive bacterial strain *Staphylococcus aureus* at the concentration of 20 mg/mL; and the least expressive, but still inhibiting growth, was also compared to *Staphylococcus aureus* at the concentration of 2.5 mg/mL. *S. lycocarpum* extract show no antibacterial activity against *Pseudomonas aeruginosa* at tested concentrations (20-0.312 mg/mL). No inhibition halo was observed in the negative control (Table 3).

Table 3: Antibacterial activity of plant extracts.

Bacterial strains	*A. falcata*		*C. regium*	
	Concentration (mg/mL)	**Diameter of inhibition halos (mm)**	**Concentration (mg/mL)**	**Diameter of inhibition halos (mm)**
Escherichia coli	20	ND	20	2.22 ± 1.1 *
	10	ND	10	1.22 ± 0.2
	5	ND	5	ND
	2.5	ND	2.5	ND
	1.25	ND	1.25	ND
	0.625	ND	0.625	ND
	0.312	ND	0.312	ND
Klebsiella pneumoniae	20	ND	20	ND
	10	ND	10	ND
	5	ND	5	ND
	2.5	2.17 ± 0.4 *	2.5	ND
	1.25	ND	1.25	ND
	0.625	ND	0.625	ND
	0.312	ND	0.312	ND
Pseudomonas aeruginosa	20	ND	20	ND
	10	1.27 ± 0.3 *	10	ND
	5	ND	5	ND
	2.5	ND	2.5	ND
	1.25	ND	1.25	ND
	0.625	ND	0.625	ND
	0.312	ND	0.312	ND
Staphylococcus aureus	20	ND	20	1.57 ± 0.1
	10	ND	10	ND
	5	ND	5	ND
	2.5	ND	2.5	ND
	1.25	ND	1.25	ND
	0.625	ND	0.625	ND
	0.312	ND	0.312	ND
Cont.				
Bacterial strains	***S. adstringens***		***S. lycocarpum***	
	Concentration (mg/mL)	**Diameter of inhibition halos (mm)**	**Concentration (mg/mL)**	**Diameter of inhibition halos (mm)**
Escherichia coli	20	1.72 ± 1.2	20	2.14 ± 0.9
	10	10.53 ± 1.2 *	10	2.1 ± 0.8
	5	ND	5	2.17 ± 1.2 *
	2.5	ND	2.5	ND
	1.25	ND	1.25	ND
	0.625	ND	0.625	ND
	0.312	ND	0.312	ND
Klebsiella pneumoniae	20	3.55 ± 1.6 *	20	ND
	10	2.7 ± 1.1	10	2.33 ± 0.7 *

	5	ND	5	2.3 ± 0.7
	2.5	ND	2.5	2.11 ± 0.8
	1.25	ND	1.25	ND
	0.625	ND	0.625	ND
	0.312	ND	0.312	ND
Pseudomonas aeruginosa	20	0.8 ± 0.2	20	ND
	10	1.22 ± 0.5 *	10	ND
	5	0.8 ± 0.1	5	ND
	2.5	ND	2.5	ND
	1.25	ND	1.25	ND
	0.625	ND	0.625	ND
	0.312	ND	0.312	ND
Staphylococcus aureus	20	2.85 ± 1.97	20	2.72 ± 1.01 *
	10	3.64 ± 1.78 *	10	2.71 ± 1.01
	5	1.16 ± 0.6	5	2.62 ± 0.7
	2.5	1.13 ± 0.2	2.5	1.14 ± 0.8
	1.25	ND	1.25	ND
	0.625	ND	0.625	ND
	0.312	ND	0.312	ND

ND = Not detectable. *P<0.05. One-way ANOVA and Bonferroni post-tests.

4. Discussion

The application of qualitative methods such as phytochemical prospecting is relevant in the health sciences due it allows low-cost screening of secondary metabolites from a plant (Matos, 1997) [11], especially when performed on potential medicinal species poorly studied nationally and/or regionally, and present in biomes of substantial interest like the Brazilian Cerrado biome. Phytochemical tests performed on the crude extracts in this study revealed the presence of several secondary metabolites groups that can contribute to the identification of chemical markers, which are indispensable for the quality and integrity tests for possible herbal formulations, as recommended by RDC No. 26 of May 13, 2014 (ANVISA, 2014) [14]. These characteristic marker substances make the raw material known, being a key factor for the standardization of herbal medicines, especially due to the great chemical variability of the plants used as medicinal products by the population (Vilegas; Cardoso; Quevedo, 2009) [15].

The result of phytochemical prospecting of secondary and constituent metabolites from the extracts of the stem of *A. falcata*, tuberous roots of *C. regium* and leaves of *S. lycocarpum* and *S. adstringens* demonstrated the presence of alkaloids, anthraquinones, catechins, coumarins, steroids and triterpenoids, phenols and tannins, flavonoids, anthocyanins and anthocyanidins, flavonols, flavonones, flavanonols and xanthones, quinones and saponins. These chemical constituents can mainly account for the possible biological activity (Ahmed *et al.*, 2017) [16]. Thus, it is important to highlight the main biological and pharmacological properties described in the literature for alkaloids, with antimalarial, anti-inflammatory, anti-diabetic, anti-rheumatic and antipyretic activities (Afewerki *et al.*, 2019; Novanna; Ethiraj; Kannadasan, 2019; Ur Rashid *et al.*, 2019) [17, 18, 19]; anthraquinones present laxative, antibacterial, antifungal, antiviral and antitumor actions (Malik & Müller, 2016; Li & Jiang, 2018) [20, 21]; catechins that belong to a group of polyphenols and present series of biological activities, such as anticarcinogenic, anti-inflammatory, antioxidant, chemoprotective and thermogenic (Schmitz *et al.*, 2005) [22]; coumarins that are described for demonstrating anticoagulant, anti-inflammatory, antifungal, antineoplastic activity and against degenerative diseases such as Alzheimer's and Parkinsonism (Jameel *et al.*, 2016; Stefanachi *et al.*, 2018; Prusty & Kumar, 2019) [23, 24, 25]; steroids perform anti-inflammatory and analgesic activities (Awang *et al.*, 2012) [26], and triterpenoids antimicrobial, antioxidant and photosensitizing activities (Souza *et al.*, 2013) [27]; phenolic compounds and tannins have an antioxidant capacity to neutralize the activity of free radicals (EROs) generated in the body, with associations with several chronic-degenerative diseases such as neoplasms, diabetes and inflammatory processes. Phenols also have the property of reducing the prevalence of cardiovascular diseases (Rocha *et al.*, 2011) [28]; tannins exert antioxidant and antimicrobial activities (antibacterial, antifungal and antiprotozoal) (Ekambaram; Perumal; Balakrishnan, 2016; Ogawa & Yazaki, 2018) [29, 30], stimulate tissue regeneration in minor ulcerations and burns, act on enzymatic and protein regulation, and stimulate phagocytic cells (Ashok & Upadhyaya, 2012; Leal *et al.*, 2015) [31, 32]; flavonoids, anthocyanins, anthocyanidins, flavonols, flavonones, flavanonols and xanthones are highlighted for their antioxidant, antiproliferative and anti-inflammatory activities (Verma & Trehan, 2013) [33], anti-ulcerogenic and antimicrobial (Cushnie & Lamb, 2005) [34], antiallergic, hepatoprotective, anticoagulant, antiviral and antineoplastic (Karabin *et al.*, 2015) [35]; quinones are capable of exerting antimicrobial and antitumor activities (Silva; Vítor; Souza, 2003; Glorieux & Buc Calderon, 2019) [36, 37]; and saponins exert antibacterial, antifungal, antiparasitic and anti-inflammatory activities (Güçlü-Ustündağ & Mazza, 2007; Saxena *et al.*, 2013; Moses; Papadopoulou; Osbourn, 2014) [38, 39, 40].

Anadenanthera genus has a wide variety of synonyms reported in the literature, as well as several popular and indigenous names (Weber *et al.*, 2011) [41]. Stems of species like *Anadenanthera falcata* are reported to exert anti-inflammatory, antioxidant and antimicrobial activity, based on the identification of steroids, flavonoids and tannins (Monteiro *et al.*, 2006; Svetaz *et al.*, 2010) [42, 43].

Solon *et al.*, (2012) [44] identified the following secondary metabolites from a hydroethanolic extract from the root of *C. regium*: ellagic acid, gallic acid, dihydrokaempferol-3-O-β-glucopyranoside, pinoresinol, dihydrokaempferol, excelsin, dihydrokaempferol-3-O-β- (6 "-galloyl) glucopyranoside, coclospermin A and B. Castro and collaborators (2004) [45] identified flavones naringenin and aromadendrin, 1-hydroxytetradecanone-3, 3-O-glycosyl dihydrokaempferol and flavonoids. And, Brum *et al.*, (1997) [46] studied the composition of the essential oil extracted from the root of *C.*

regium with a yield about 0.25%, and observed the following constituents: β-selinene, β-elemene, transcaryophyllene, α-pinene, α-humulene, α-selinene and δ-cadinene.

S. adstringens has been submitted to phytochemical analyzes by several authors. Scalbert (1991) [47] demonstrated the antibacterial potential of *S. adstringens* attributed to tannins, since tannins are toxic to fungi and bacteria, due to their properties in inhibiting the action of extracellular enzymes and oxidative phosphorylation, in addition to mechanisms involving substrate and iron deprivation, which are necessary for these microorganisms' survival. Ishida *et al.*, (2006) [48] evaluated the antifungal activity of *S. adstringens* fractions against *Candida albicans* and showed the plant interfered in the growth and in the virulence factors, as well as presenting low toxicity to mammalian cells. According to these authors, the antifungal effect is due to the presence of proanthocyanidins, which include several flavan-3-ools, such as prodelphinidin and prohorbinetinidines. In another study, Thomazi; Bertolin; Pinto (2010) [49] evaluated the antibacterial activity from the bark and leaf of *S. adstringens*, verifying antimicrobial potential against several bacterial strains: *Proteus mirabilis*, *Enterobacter sp*, *Staphylococcus saprophyticus*, *Staphylococcus aureus*, *Staphylococcus sp.*, *Escherichia coli*, *Citrobacter sp.*, *Enterobacter aggomerans*, *Pseudomonas aeroginosa* and *Klebsiella oxytoca*.

Phytochemical analysis of *S. lycocarpum* carried out by several authors reveal an abundance of compounds with pharmacological properties that justify studies on the use of the plant against different pathologies. Miranda *et al.*, (2013) [50] demonstrated the abundant presence of two steroidal glycoalkaloids in the extract of *S. lycocarpum* fruits, solasonine and solamargine. Studies by Morais and collaborators (2015) [51] indicated that caffeic acid and chlorogenic acid were the main phenolic compounds present in an ethanolic extract of the ripe fruits of *S. lycocarpum*. In this study, authors also stated that the extract exerts antibacterial activity against several Gram-positive strains, such as *Streptococcus mutans*, *Streptococcus pyogenes*, *Staphylococcus aureus* and *Enterococcus faecalis*. Jimoh; Adedapo; Afolayan, (2010) [52] and Bontempo *et al.*, (2013) [53] also describe that species of *Solanum* demonstrate antibacterial properties.

The knowledge about the groups of secondary metabolites from medicinal plants, even if due to their preliminary qualification, has an important control parameter in the chemical marker, which does not disregard the importance of establishing reference profiles through the quantification of these markers in the different geographical locations of the vast Brazilian territory. However, discrepancies in results occur in qualitative and even quantitative comparisons of secondary metabolites and this is due to aspects related to soil, climate, material collection, temperature and chemical reagents used (Silva; Miranda; Conceição, 2010) [2]. Such approaches demonstrate that the phytochemicals of the secondary metabolism may, for the same species, present variations owing to ecosystem environmental differences (Matos, 1997) [11]. This issue assumes relevance in view of the need for vegetable raw materials standardization aiming at the validation of medicinal plants locally used, the control of existing herbal medicines and the quality assured for new ones, in addition to support for research involving potential biological activities.

The biggest issue related to the evaluation of plant antimicrobial activity is the lack of uniformity in the criteria, which often leads to contradictions between the results obtained by different researchers and even for the same author using the same sample with different methods (Gobbo-Neto & Lopes, 2007) [54]. According to Wiest *et al.*, (2009) [55] there is evidence that the type of extraction directly influences solutions antibacterial efficacy. In the same vein, values of inhibitory concentrations may differ due to a series of factors such as seasonality, temperature, water availability, ultraviolet radiation, altitude, availability of nutrients, exposure to pathogens, among others (Ríos & Recio, 2005) [56].

5. Conclusion

Empirical therapeutic indications pointed out by the Goiana community indicate biological and pharmacological actions of phytochemical constituents present in the extracts from the stem of *A. falcata*, tuberous roots of *C. regium* and leaves of *S. adstringens* and *S. lycocarpum*.

Data obtained in this study direct future approaches to verify the biological activity of these plants based on the presence of relevant phytochemical constituents, such as alkaloids, anthraquinones, catechins, coumarins, steroids and triterpenoids, phenols and tannins, flavonoids, anthocyanins and anthocyanidins, flavonols, flavonones, flavanonols and xanthones, quinones and saponins with antibacterial potential. Markers characterization is indispensable for plants validation, in order to indicate safer popular uses and for tests of quality and integrity of herbal medicines, which allows a pharmacognostic control of native species from the Brazilian Cerrado biome.

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