

Article

Variability of Volatile Oils Composition, Tannins, and Phenols from *Campomanesia adamantium* (CAMBESS.) O. Berg

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ABSTRACT

Campomanesia adamantium (Myrtaceae), known as "guabiroba-do-campo", is a native Cerrado shrub popularly used as anti-inflammatory, antidiarrheal, and urinary tract antiseptics. This study aimed to evaluate the seasonal variability of total phenols and tannins and chemical compounds of the volatile oils of *C. adamantium* leaves over 12 months. The leaves and flowers were collected in Bela Vista, Goiás, Brazil. The volatile oils were obtained by hydrodistillation in a Clevenger apparatus and analyzed by Gas Chromatography-Mass Spectrometry (GC/MS). The determination of total phenols and tannins was performed by the method of Hagerman and Butler. The main constituents of the essential oil from the leaves were γ -elemene, limonene, italicene epoxide, β -funebrene, bicyclogermacrene, and linalool, and those of flower oil were sabinene, limonene, linalool, tricyclene, and methyl salicylate. The total phenols content ranged from 3.75% to 9.56% and the tannins content from 2.25% to 4.84%. The best period for collecting the leaves with the highest index of phenols and tannins is low rainfall. This work represents the first description of the seasonal variability of the essential oils, tannins, and total phenols of *C. adamantium* collected in Bela Vista.

Keywords: carobinha; cerrado; Myrtaceae; medicinal plants; essential oils.

RESUMO

Campomanesia adamantium (Myrtaceae), conhecida como "guabiroba-do-campo", é um arbusto nativo do Cerrado popularmente usado como anti-séptico anti-inflamatório, antidiarreico e do trato urinário. Os objetivos deste estudo foram: avaliar a variabilidade sazonal dos fenóis totais e taninos e compostos químicos dos óleos voláteis das folhas de *C. adamantium* em um período de 12 meses. As folhas e flores foram coletadas em Bela Vista, Goiás, Brasil. Os óleos voláteis foram obtidos por hidrodestilação em aparelho de Clevenger e analisados por Cromatografia gasosa-espectrometria de massa (CG/EM). A determinação dos fenóis totais e taninos foi realizada pelo método de Hagerman e Butler. Os principais compostos dos óleos das folhas foram γ -elemeno, limoneno, epóxido de italiceno, β -funebrene, bicyclogermacrene, linalol e das flores foram sabineno, limoneno, linalol, triciclono e salicilato de metila. O teor total de fenóis variou de 3,75% a 9,56% e o teor de taninos, de 2,25% a 4,84%. Concluiu-se que o melhor período para coleta das folhas com maior índice de fenóis e taninos é a baixa precipitação. Este trabalho representa a primeira descrição da variabilidade sazonal dos óleos essenciais, taninos e fenóis totais de *C. adamantium* coletados em Bela Vista.

Palavras-chave: carobinha; cerrado; Myrtaceae; plantas medicinais; óleos essenciais.



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1. Introduction

In Brazil, the Myrtaceae family has 23 genera and 976 species (Sobral et al. 2010), with economic potential, being many fruitful species such as guava (*Psidium guajava* L.), jabuticabeira (*Myrciaria cauliflora* Mart. O. Berg.) and the pitangueira (*Eugenia uniflora* L.) (Gressler et al. 2006). There are 14 genera and 211 species in the Cerrado (Sobral et al. 2010).

Studies of the chemical profile of Myrtaceae described the presence of flavonoids (Ferreira et al. 2006, Klafke et al. 2010, Mustafa et al. 2005, Paula et al. 2008), tannins (Markman et al. 2004, Tanaka et al. 1996, Yang et al. 2000), phloroglucinol adducts (Umehara et al. 1998), acetophenones (Yoshikawa et al. 1998), volatile oils and terpenes (Benyahia et al. 2005, Cardoso et al. 2008, Djoukeng et al. 2005, Gu et al. 2001, Osorio et al. 2006).

Campomanesia adamantium (Cambess.) O. Berg (Myrtaceae), popularly known as "guabiroba-do-campo" is a deciduous shrub 0.5-1.5 m tall native to the Cerrado (Lima et al. 2011, Lorenzi et al. 2008). *C. adamantium* has glabrescent leaves, with anomocytic and paracytic stomata, grouped between the veins. The secretory cavities are distributed along the entire length of the leaf blade, indistinctly adjacent to the abaxial and adaxial surfaces (Gomes et al. 2009). The fruits are globose, yellowish green, bacoid type. They are tasty and edible to the natural, used in the preparation of juices, jellies, liqueurs, and ice creams. They serve as food for birds of the Cerrado like jacus (*Penelope* spp.), sanhaços (*Tangara* spp.), mammals such as maned wolf (*Chrysocyon brachyurus*), field fox (*Pseudalopex vetulus*) (Kuhlmann 2012).

C. adamantium leaves are popularly used as anti-inflammatory, antidiarrheal, and antiseptic of the urinary tract, for stomach disorders (Lorenzi et al. 2008, Piva 2002), and to treat diabetes and cholesterol (Kuhlmann 2012).

Viscardi et al. (2017) identified the flavonoids 3,5,7,3',4',5'-hexahydroxy-flavonol, 3,5,7,3',4',5'-hexahydroxy-flavonol-3-O- α -L-arabinofuranoside, 3,5,7,3',4',5'-hexahydroxy-flavonol-3-O- α -L-raminopyranoside, 7-dihydroxy-5-metoxiflavanone, 6-methyl-7-hydroxy-5-metoxiflavanone, 2',4'-dihydroxy-6'-metoxichalcone, and 2',4'-dihydroxy-5'-methyl-6'-metoxichalcone. Sá et al. (2018) isolated and identified estictano-3,22-diol from the hexane fraction and valoneic acid and gallic acid from the aqueous fraction. The volatile oils extracted from the *C. adamantium* leaves during the flowering and fruiting period showed high activity against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Candida albicans* and moderate activity against *Escherichia coli* (Coutinho et al. 2009). Pavan et al. (2009) described the antimicrobial activity of the fractions from the ethyl acetate extract of the *C. adamantium* fruits containing flavonoids against *Mycobacterium tuberculosis*. Cardoso et al. (2010) verified the antimicrobial activity of the extract and hexane fraction of *C. adamantium* fruits against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Salmonella setubal*, *Saccharomyces cerevisiae*, and *Candida albicans*. Sá et al. (2018) observed good antimicrobial activity of the crude extract and leaf fractions against Gram-positive bacteria and fungi, leaf volatile oils and flowers against *Listeria monocytogenes*, *Trichophyton mentagrophytes* and valoneic acid against *Cryptococcus neoformans*, *Candida tropicalis*, *Candida krusei*, *Candida parapsilosis*, *T. mentagrophytes*, and *Trichophyton rubrum*.

Coutinho et al. (2008a) verified *in vitro* antioxidant activity of the hexane, chloroform, and ethanol extracts from *C. adamantium* leaves. Ferreira et al. (2013) found that the ethyl acetate and aqueous fractions and flavonoids myricetin and myricetin isolated from the hexane fraction from *C. adamantium* leaves showed anti-nociceptive and anti-inflammatory effects. Pascoal et al. (2014) described that a chalcone named cardamonin isolated from the ethanol extract from *C. adamantium* leaves inhibited the growth of prostate cancer cells. Viscardi et al. (2017) verified to anti-inflammatory and antihyperalgesic activities of the pulp of the microencapsulated fruits of *C. adamantium* in rats.



The quality of vegetable raw materials plays a fundamental role in obtaining products with constant chemical composition and therapeutic properties that can be reproduced. According to Gobbo-Neto and Lopes (2007) Barros et al. (2009) the secondary metabolism of plants can vary considerably and not maintain its constancy throughout the year.

This study aimed to evaluate the seasonal variability of the total phenols, tannins, and, chemical compounds of the volatile oils from *C. adamantium* leaves over 12 months.

2. Material and methods

2.1. Plant material

Campomanesia adamantium (Cambess.) O. Berg leaves were collected monthly (300 g) from ten different plants, from February 2015 to January 2016, and flowers in October 2015 in Bela Vista, Goiás, Brazil (17° 02' 01.1'' S; 48° 49' 00.3'' W; at an elevation of 847 m above sea level). Prof. Dr. José Realino de Paula identified the plant material and a voucher specimen was deposited at the Herbarium of Federal University of Goiás under the code number UFG-243832.

Climatic data for the period were obtained from the Meteorological Institute (INMET, 2017).

2.2. Volatile oils of the leaves and flowers

For the extraction of the volatile oils, leaves and flowers (100 g) were dried at room temperature for three days, triturated using a commercial crusher (Skymen, LS-08MB-N) immediately before the extraction of the volatile oil, avoiding loss by volatilization, and submitted to hydrodistillation in a Clevenger-type apparatus for 2 h. After drying with anhydrous Na₂SO₄, the oils were stored in glass vials at a temperature of -18 °C until further analysis. The volume of the essential oils was measured in the graduated tube of the apparatus and was calculated as a percentage of the initial amount of dry plant material used in the extraction. Each experiment was performed in triplicate.

Essentials oils from leaves (EOl) and flowers (EOfl) were analyzed using a Shimadzu GC/MS-QP5050A fitted with a fused silica SBP-5 (30 m × 0.25 mm I.D.; 0.25µm film thickness) capillary column (composed of 5% phenylmethylpolysiloxane). The following temperature program was used: the temperature was raised from 60-240 °C at 3 °C/min and then to 280 °C at 10 °C/min, ending with 10 min at 280 °C. The carrier gas (Helium) had a flow rate of 1 ml/min, and the split mode had a ratio of 1:20. The injection port was set at 225 °C.

The significant operating parameters for the quadrupole mass spectrometer were as follows: interface temperature, 240 °C; electron impact ionization at 70 eV with a scan mass range of 40-50 *m/z* at a sampling rate of 1 scan/s.

Constituents were identified by an electronic search using digital libraries of mass spectral data (NIST, 1998), comparison of the retention indices of the constituents (Van Den Dool and Kratz 1963) to those of C₈–C₃₂ n-alkanes, and comparison of the mass spectra with literature data (Adams 2007).

2.3. Extraction and dosing of total phenols (TP) and tannins (PP)

The determination of total phenols and tannins of the leaves of *C. adamantium* collected monthly was carried out according to the methodology described by Hagerman and Butler (Mole and Waterman, 1987). The powder of the leaves (0.75 g) was extracted with distilled water (150 mL). The mixture was heated to boiling, after being kept in the water bath at between 80 and 90 °C for 30 min. The contents of the flask were transferred to a 250 mL volumetric flask and the volume was made up of distilled water. The extract was filtered through



qualitative filter paper, with the first 50 mL discarded. The aqueous extract obtained was used for quantification of total phenols (TP) and for the tannins protein precipitation assay (PP).

For total phenolics assay (TP), ferric chloride was added to aqueous extract under alkaline conditions to result in a colored complex with phenolic compounds (read at 510 nm). The standard curves were prepared with tannic acid at the dilutions: 0.10, 0.15, 0.20, 0.25, and 0.30 mg/mL.

For the tannins protein precipitation assay (PP), the aqueous extracts were precipitated with Bovine Serum Albumine (BSA) in 0.2 M acetate buffer (pH 4.9) and after centrifugation, the precipitated (containing tannins) was dissolved in sodium dodecyl sulfate/triethanolamine solution, then ferric chloride was added and tannins were complexed (read at 510 nm). The standard curves were prepared with tannic acid at the dilutions: 0.10, 0.20, 0.30, 0.40, and 0.50 mg/mL.

All experiments were performed in triplicate.

2.4. Statistical analyses

The data were analyzed using the statistical software Statistica (Stat Soft 2004). To assess the chemical variability, principal component analysis (PCA) was applied to examine the interrelationships between the chemical constituents of the essential oils from leaves collected in different months. A hierarchical cluster analysis (HCA) was used to study the similarity of samples based on the distribution of the major constituents of the essential oils. Hierarchical clustering was performed according to the method of Ward's variance-minimizing method (Ward 1963). To validate the classification proposed by HCA, a canonic discriminant analysis (DCA) was employed. The mean values of TP and TT in each cluster proposed by the HCA were compared using Student's t-test, and $p < 0.05$ was considered statistically significant.

For the comparison of the two seasons that occur in the Cerrado biome, the Student's t-test was used, using the major compounds present in the essential oil of *C. adamantium*. Past software 3.20 was used for this comparison between the two groups (Hammer & Harper 2001).

3. Results and discussion

During the collection period the months of highest rain-fall were February/ 2015 (155.1 mm), March/2015 (156.2 mm), November/2015 (354.8 mm), December/2015 (207.7 mm), January/ 2016 (484.8 mm); with average temperatures ranging from 20.0°C to 33.9°C. The months with less rainfall were April /2015 (1.3 mm), June/2015 (0.0 mm), July/2015 (2.7 mm), August/2015 (3.6 mm), September /2015 (30.4 mm), October/2015 (18.2 mm), temperatures ranging from 16.7°C to 36.7°C (Table 1).

3.1. Volatile oils

The yields of the volatile oils from *C. adamantium* leaves ranged from 0.5 to 2.5% (v / w) and of the flower was 1.5% (Table 2). In volatile oils of the leaves were identified 1.5 to 40.6% of monoterpenes, 2.2 to 13.6% of oxygenated monoterpenes, 31.5 to 63.3% of sesquiterpenes, 2.0 to 23.2% of oxygenated sesquiterpenes and 0.7 to 1.4% other compounds. In volatile oils of the flowers were identified 53.83% monoterpenes, 14.6% oxygenated monoterpenes, 6.1% sesquiterpenes, 9.4% oxygenated sesquiterpenes and 8.7% other compounds (Table 2).

The major compounds of leaf oils were γ -elemene (ranging from 3.8 to 30.4%), limonene (1.5 to 27.7%), italicene epoxide (0.9 to 18.6%), β -funebrene (5.5 to 12.9%), bicyclogermacrene (0.5 to 8.9%), and linalool (0.4 to 8.7%). The major compounds of the flowers were sabinene (20.5%), limonene (19.3%), linalool (10%), tricyclene (9.1%), and methyl salicylate (8.7%).



The results obtained from the PCA and cluster analysis showed the existence of chemical variability among samples of oils obtained from *C. adamantium* leaves (Figure 1). Figure 2 indicates that the relative position of the axis 2D originated in the PCA. This analysis suggests that cluster I is discriminated by compounds cis-eudesma-6,11-diene and γ -elemene (volatile oils from leaves collected in November, December, January, February, March, and April). The samples present in cluster I were characterized by a period with higher levels of rainfall. Cluster II (volatile oils from leaves collected in June, July, August, September, and October) (Figure 2) suggests that linalool, tricyclene, italicene epoxyde, α -guaiene, β -funebrene are the compound capable of discriminating this group characterized in samples collected in months of low rainfall. The results of the canonical discriminant analysis indicate that the classification proposed by the PCA and HCA was appropriate for the classification of samples as the chemical profile of volatile oils.

Canonical discriminant analysis was performed to predict the grouping of the cluster analysis, and two predictive variables were employed: tricyclene and γ -elemene the two discriminant functions retain 100% of well - classification in the original clusters by a cross-validation approach. Thus, the canonic discriminant analysis revealed that the classification proposed and the variables employed are suitable to show that the findings of the HCA and the PCA were consistent (Table 3).

For the comparison of the two seasons observed in the Cerrado biome (hot and dry-April to October and rainy-November to March), the following compounds were selected for comparison with the two periods: α -pinene, limonene, linalool, β -funebrene, γ -elemene, italicene epoxide, and aromadendrene. The two seasons presented significant differences only concerning the components: linalool (3.7% for hot and dry months and 1.1% for rainy season, $p < 0.1$), italicene epoxide (7.7% for the hot and dry months and 3.1% for the rainy season $p < 0.1$) aromadendrene (1.61% for hot and dry months and 1.12% for the rainy season, $p < 0.05$). Coutinho et al. (2008, 2009) verified as major compounds in the essential oil of *C. adamantium* leaves collected in Mato Grosso do Sul State, Brazil, the limonene at the period of flowering, and the bicyclogermacrene in the period of fruiting. According to Stefanello et al. (2008), the oil from the leaves of *C. adamantium* was characterized by a predominance of sesquiterpenes (59.9%), mainly of the germacrene group, and significant amounts of monoterpenes (28.7%). Aromatic compounds, represented by methyl salicylate and eugenol, were found as minor constituents. The main components were geraniol (18.1%), spathulenol (7.8%), and globulol (5.6%). According to Oliveira et al. (2017) the major constituents identified in essential oils from *C. adamantium* leaves were spathulenol (19.27%), germacrene-B (18.27%), and β -caryophyllene oxide (12.37%). Sá et al. (2018) verified as major compounds of the volatile oils of *C. adamantium* leaves collected in October 2012 in Goiás: the verbenene, β -funebrene, limonene, α -guaiene, and linalool. According to Gobbo-Neto and Lopes (2007) and Lima et al. (2003) the volatile oil chemical composition is genetically determined and many abiotic factors. Light, temperature, seasonality, nutrition, rainfall index, herbivory, and the circadian cycle can significantly change the production of secondary metabolites. Barros et al. (2009) state that temperature can influence terpenoids by affecting the enzymatic activities in some species and, consequently, interfering with the biosynthesis of some secondary metabolites. According Lee and Ding (2016) microclimatic factors such as temperature, rainfall distribution, and geographical features, especially altitude contribute to the differences in the chemotype of certain essential oil bearing plants. The type and nature of the constituents and their concentration levels are important attributes, particularly in terms of biological activities of the essential oils (Batish et al. 2008). Climatic factors such as temperature and precipitation have also been taken into account in the case of *Porcelia macrocarpa*. Maximum essential oil yield was obtained in June with lower temperature, while minimum essential oil was obtained in December/January when higher temperature and precipitation were recorded. Silva et al. (2013) suggested the inverse proportional relation between essential oil yields and the



temperature and precipitation pattern. Essential oil variations due to rainfall and temperature were also reported on lavender (*Lavandula angustifolia*) during the flowering period (Hassiotis et al. 2014). The essential oil quality of lavender is characterized by the presence of its major compounds, linalool, and linalyl acetate. A relative amount of linalool dropped after rainfall followed by several days of low temperatures resulting in lower quality of lavender oil. However, the linalool content increased after 15 days, therefore restoring the oil quality. On the contrary, compounds such as α -terpineol, borneol, and lavandulyl acetate are characterized by enhanced production after rainfall. The different compositions of the *C. adamantium* essential oils may suggest high chemical variability, probably related to variation associated with rainfall and with the behavior of specimens throughout the year.

Limberger et al. (2001), studied the volatile oils of the leaves of four species of the genus, *Campomanesia aurea* O. Berg, *Campomanesia guazumifolia* (Cambess.) O. Berg, *Campomanesia rhombea* O. Berg and *Campomanesia xanthocarpa* O. Berg, verified, in all the samples, sesquiterpenes, among them, spathulenol, caryophyllene oxide, bicyclogermacrene and (E)-pylidol. Other species of the genus, *Campomanesia phaea* (O. Berg) Landrum, *Campomanesia guaviroba* (DC.), *Campomanesia sessiliflora* O. Berg, presented as major compounds of volatile leaf oil, caryophyllene oxide (Adati & Ferro, 2006), myrtenal (Pascoal et al. 2011) and bicyclogermacrene (Cardoso 2010). The yield of leaf essential oil, found by these authors, was approximately 0.5%, and in the present work ranged from 0.5 to 2.5%.

3.3. Dosing of total phenols (FT) and tannins (TT)

The total phenol content in the powder of *C. adamantium* leaves ranged from 3.75% to 9.56% and tannin content from 2.25% to 4.84% (Table 4). There was an increase in total phenols between April to September and a reduction in their contents from October (Figure 3).

There was a significant difference at the 5% level between the total phenols and tannin contents between the samples of the two clusters, verifying an increase in the contents in the period of low rainfall. According to Sá et al. (2018) the month's low rainfall (August to October) is the period of flowering and fruiting of this specie. Coutinho et al. (2010) found through HPLC analysis an increase in chalcones and flavanones content in early spring (hot and dry climate) in *C. adamantium* leaves collected in the city-Bela Vista, MS. This increase, according to the authors, may be related to the accumulation of flavonoids on the leaf surface as protection against UV rays and/or even as insect defense.

In conclusion, based on the biological activities of *C. adamantium* researched and many related to phenolic compounds such as flavonoids, it is suggested through the present study that the best period for collecting the leaves with the highest index of these compounds is that of low rainfall. It was verified the existence of two clusters in the volatile oils of the leaves, one in the rainy period and another in the dry period. This work represents the first description of the seasonal variability of the essential oils, tannins, and flavonoids of *C. adamantium* leaves collected in Bela Vista, Goiás State, Brazil.

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References

- Adams RP. 2007. *Identification of essential oil components by Gas Chromatography/Mass Spectrometry*. 4. ed., Allured Publishing Corporation, Illinois, 804 pp.
- Adati RT, Ferro VO 2006. Volatile oil constituents of *Campomanesia phaea* (O. Berg) Landrum. (Myrtaceae). *J Essent Oil Res* 18 (6): 691-692.
- Barros FMC, Zambarda EO, Heinzmann BM, Carlos A, Mallmann CA 2009. Variabilidade sazonal e biossíntese de terpenóides presentes no óleo essencial de *Lippia alba* (Mill.) N. E. Brown (Verbenaceae). *Quim Nova* 32 (4): 861-867.
- Batish DR, Singh HP, Kohli RK, Kaur S 2008. Eucalyptus essential oil as a natural pesticide. *For Ecol Manag* 256: 2166-2174.
- Benyahia S, Benayache S, Benayache F, León F, Quintana J, Lopez M, Hernández JC, Estévez F, Bermejo J 2005. Cladocalol a pentacyclic 28- nor-triterpene from *Eucalyptus cladocalyx* with cytotoxic activity. *Phytochem* 66: 627-632.
- Brasil. 2010. *Farmacopeia Brasileira*. 5ª ed., Atheneu, São Paulo, 546 pp.
- Cardoso CAL, Silva JRM, Kataoka VMF, Brum CS, Poppi NR 2008. Avaliação da atividade antioxidante, toxicidade e composição química por CG-EM do extrato hexânico das folhas de *Campomanesia pubescens*. *Rev Cien Farm Bas Aplic* 29: 297-301.
- Cardoso C, Salmazzo G, Honda NK, Prates CB, Vieira MC, Coelho RG 2010. Antimicrobial activity of the extracts and fractions of hexanic fruits of *Campomanesia* species (Myrtaceae). *J Med Food* 13 (5): 1273-1276.
- Coutinho ID, Coelho RG, Kataoka VMF, Honda NK, Silva JRM, Vilegas W, Cardoso CAL. 2008a. Determination of phenolic compounds and evaluation of antioxidant capacity of *Campomanesia adamantium* leaves. *Eclét Quím* 33 (4): 53-60.
- Coutinho ID, Poppi NR, Cardoso C. 2008b. Identification of the volatile compounds of leaves and flowers in guavira (*Campomanesia adamantium* O. Berg.). *J Essent Oil* 20 (5): 405-407.
- Coutinho ID, Cardoso CAL, Ré-poppi, Melo AM, Vieira MC, Honda NK, Coelho RG. 2009. Gas Chromatography-Mass Spectrometry (GC-MS) and evaluation of antioxidant and antimicrobial activities of essential oil of *Campomanesia adamantium* (Cambess.) O. Berg (Guavira). *Braz J Pharm Sci* 45 (4): 767-776.
- Coutinho ID, Kataoka VMF, Honda NK, Coelho RG, Vieira MC, Claudia AL, Cardoso CAL 2010. Influência da variação sazonal nos teores de flavonóides e atividade antioxidante das folhas de *Campomanesia adamantium* (Cambess.) O. Berg, Myrtaceae. *Braz J Pharmacog* 20(3): 322-327.
- Djoukeng JD, Abou-mansour E, Tabacchi R, Tapondjou AL, Bouda H, Lontsi D. 2005. Antibacterial triterpenes from *Syzygium guineense* (Myrtaceae). *J Ethnopharmacol* 101: 283-286.



- Ferreira ACF, Neto JC, Silva ACM, Kuster RM, CARVALHO DP 2006. Inhibition of thyroid peroxidase by *Myrcia uniflora* flavonoids. *Chem Res Toxicol* 19 (3): 351-355.
- Ferreira LC, Grabe-Guimarães A, Michel MC, Guimarães RG, Rezende SA. 2013. Anti-inflammatory and antinociceptive activities of *Campomanesia adamantium*. *J Ethnopharmacol* 145 (1): 100-108.
- Gobbo-Neto L, Lopes NP 2007. Plantas medicinais: fatores de influência no conteúdo de metabólitos secundários. *Quim Nova* 30 (2): 374-381.
- Gomes SM, Somavilla NSDN, Gomes-Bezerra KM, Miranda SC, Carvalho OS, Graciano-Ribeiro D 2009. Anatomia foliar de espécies de Myrtaceae: contribuições à taxonomia e filogenia. *Acta Bot Bras* 23(1): 223-238.
- Gressler E, Pizo MA, Morellato LPC 2006. Polinização e dispersão de sementes em Myrtaceae do Brasil. *Braz J Bot* 29 (4): 509-530.
- Gu JQ, Park EJ, Luyengi L, Hawthorne ME, Mehta RG, Farnsworth N R, Pezzuto JM, Kinghorn AD 2001. Constituents of *Eugenia sandwicensis* with potential cancer chemopreventive activity. *Phytochem* 58: 121-127.
- Hammer Ø, Harper DAT, Ryan PD 2001. PAST: Paleontological Statistics Software package for education and data analysis. *Palaeontol Electr* 4:1-9.
- Hassiotis CN, Ntana F, Lazari DM, Poullos S, Vlachonassios KE 2014. Environmental and developmental factors affect essential oil production and quality of *Lavandula angustifolia* during flowering period. *Ind Crop Prod* 62: 359-366.
- INMET. 2017. *Instituto Nacional de Meteorologia. Ministério da agricultura, pecuária e abastecimento*. [updated 2017 May]. Available from: <http://www.inmet.gov.br/portal/>.
- Klafke JZ, Silva MA, Panigas TF, Belli KC, Oliveira MF, Barichello MM, Rigo FK, Rossato MF, Santos ARS, Pizzolatti MG, Ferreira J, Vicili PR 2010. Effects of *Campomanesia xanthocarpa* on biochemical, hematological and oxidative stress parameters in hypercholesterolemic patients. *J Ethnopharmacol* 127: 299-305.
- Kuhlmann M 2012. *Frutos e sementes do Cerrado atrativos para a fauna*. Ed. Rede de Sementes do Cerrado, Brasília, 360 pp.
- Lee YL, Ding P 2016. Production of essential oil in plants: Ontogeny, secretory structures and seasonal variations. *PJSRR* 2(1): 1-10
- Lima HRP, Kaplan MAC, Cruz AVM 2003. Influência dos fatores abióticos na produção e variabilidade de terpenoides em plantas. *Floresta e Ambiente* 10:71-77.
- Lima DF, Goldenberg R, Sobral M 2011. O gênero *Campomanesia* (Myrtaceae) no estado do Paraná, Brasil. *Rodriguésia* 62 (3): 683-693.
- Limberger RP, Apel MA, Sobral M, Menut LC 2001. Chemical composition of essential oils from some *Campomanesia* Species (Myrtaceae). *J Essent Oil Res* 13 (2): 113-115.



Lorenzi H, Lacerda M, Bacheret L 2008. *Frutas brasileiras e exóticas cultivadas*. Ed. Instituto Plantarum, São Paulo, 640 pp.

Markman BEO, Bacchi EM, Kato ETM 2004. Antiulcerogenic effects of *Campomanesia xanthocarpa*. *J Ethnopharmacol* 94 (1): 55-7.

Mole S, Waterman PG 1987. A critical analysis of techniques for measuring tannins in ecological studies. *Oecologia* 72: 137-147.

Mustafa K, Perry NB, Weavers RT 2005. Lipophilic C-methyl flavonoids with no B-ring oxygenation in *Metrosideros* species (Myrtaceae). *Biochem Syst Ecol* 33: 1049-1059.

NIST 1998. *National Institute of Standards and Technology, PC Version of the NIST/EPA/NIH Mass Spectral Database*. U.S. Department of Commerce, Gaithersburg.

Oliveira JDD, Alves DKM, Miranda MLD, Alves JM, Xavier MN, Casal CDM, Alves CCF 2017. Chemical composition of essential oil extracted from leaves of *Campomanesia adamantium* subjected to different hydrodistillation times. *Ciência Rural* 47(1):1-7.

Osório C, Alarcon M, Moreno C, Bonilla A, Barrios J, Garzón C, DUQUE C 2006. Characterization of odor-active volatiles in champa (*Campomanesia lineatifolia* R. & P.). *J Agricult Food Chem* 54: 509-516.

Pascoal ACRF, Lourenco CC, Sodek L, Tamashiro JY, Franchi JRGC, Nowill AE, Stefanello MEA, Salvador MJ 2011. Essential oil from the leaves of *Campomanesia guaviroba* (DC.) Kiaersk. (Myrtaceae): Chemical composition, antioxidant and cytotoxic activity. *J Essent Oil Res* 23 (5): 34-37.

Pascoal ACRF, Ehrenfried CA, Lopez BGC, Araujo TM, Pascoal VDB, Gilioli R, Anhê GF, Ruiz ALTG, Carvalho JE, Stefanello MEA, Salvador MJ 2014. Antiproliferative activity and induction of apoptosis in PC-3 cells by the chalcone cardamonin from *Campomanesia adamantium* (Myrtaceae) in a bioactivity-guided study. *Molecules* 19 (2): 1843-1855.

Paula JAM, Paula JR, Bara MTF, Rezende MH, Ferreira HD 2008. Estudo farmacognóstico das folhas de *Pimenta pseudocaryophyllus* (Gomes) L.R. Landrum Myrtaceae. *Rev Bras Farmacogn* 18: 265-278.

Pavan FR, Leite CQF, Coelho RG, Coutinho ID, Honda NK, Cardoso CAL, Vilegas W, Leite SRA, Sato DN 2009. Evaluation of anti-*Mycobacterium tuberculosis* activity of *Campomanesia adamantium* (Myrtaceae). *Quim Nova* 32 (5): 1222-1226.

Piva MG 2002. *O Caminho das plantas medicinais: estudo etnobotânico*. v 1, Editora Mondrian, Rio de Janeiro, 313 pp.

Sobral M, Proença C, Souza M, Mazine F, Lucas E 2010. Myrtaceae. Lista de espécies da flora do Brasil. [updated 2010 Sep 23]. Available from: <http://www.floradobrasil.jbrj.gov.br/>.

STATSOFT INC. 2004. *Statistica: data analysis software system, version 7*. Tulsa. Available from: <http://www.statsoft.com/>



- Sá S, Chaul LT, Alves VF, Fiuza TS, Leonice LMF, Vaz BG, Ferri PH, Borges LL, Paula JR 2018. Phytochemistry and antimicrobial activity of *Campomanesia adamantium*. *Rev Bras Farmacog* 28: 303–311.
- Stefanello MEA, Cervi AC, Wisniewski JrA, Simionatto EL 2008. Essential Oil Composition of *Campomanesia adamantium* (Camb) O. Berg. *J Essent Oil Res* 20(5): 424-425.
- Tanaka T, Orii Y, Nonaka G, Nishioka I, Kouno I 1996. Syzyginins A and B, two ellagitannins from *Syzygium aromaticum*. *Phytochem* 43: 1345-1348.
- Silva E, Soares M, Mariane B, Vallim M, Pascon R, Sartorelli P, Lago J 2013. The seasonal variation of the chemical composition of essential oils from *Porcelia macrocarpa* R. E. Fries (Annonaceae) and their antimicrobial activity. *Molecules* 18: 13574-13587.
- Umehara K, Singh IP, Etoh H, Takasaki M, Konoshima T 1998. Five phloroglucinol-monoterpene adducts from *Eucalyptus grandis*. *Phytochem* 49(6): 1699-1704.
- Van Den Dool H, Kratz PD 1963. Generalization of the Retention Index system including linear temperature programmed gas-liquid partition chromatography. *J Chromatogr* 11: 463-471.
- Viscardi DZ, Oliveira VS, Arrigoa JS, Piccinelli AC, Cardoso CAL, Maldonadee IR, Kassuyac CAL, Sanjinez-Argandona EJ 2017. Anti-inflammatory, and antinociceptive effects of *Campomanesia adamantium* microencapsulated pulp. *Rev Bras Farmacog* 27: 220–227.
- Ward JH 1963. Hierarchical grouping to optimize an objective function. *J Am Stat Assoc.* 58: 66-103.
- Yang LL, Lee CY, Yen KY 2000. Induction of apoptosis by hydrolyzable tannins from *Eugenia jambos* L. on human leukemia cells. *Cancer Lett* 157: 65-75.
- Yoshikawa M, Shimada H, Nishida N, Toguchida I, Yamahara J, Matsuda H 1998. Antidiabetic principles of natural medicines. II. Aldose reductase and alpha-glucosidase inhibitors from Brazilian natural medicine, the leaves of *Myrcia multiflora* DC. (Myrtaceae): Structures of myrciacitrins I and II and myrciaphenones A and B. *Chem Pharm Bul* 46(1):113-119.


Table 1 Climate information of collection period of *C. adamantium*.

Station	Date	Rainfall total	Average maximum temperature (°C)	Average minimum temperature (°C)	Relative humidity
83423	02/20/2015	155.1	33.2	21.0	68.9
83423	03/31/2015	156.2	32.3	21.2	67.8
83423	04/30/2015	1.3	33.4	20.2	50.3
83423	05/31/2015	70.7	29.7	18.4	66.0
83423	06/30/2015	0	30.2	17.0	56.3
83423	07/31/2015	2.7	31.4	16.7	50.9
83423	08/31/2015	3.6	33.3	17.5	38.4
83423	09/30/2015	30.4	30.4	20.3	42.5
83423	10/31/2015	18.2	36.7	22.1	43.5
83423	11/30/2015	354.8	33.9	21.0	63.9
83423	12/31/2015	207.7	33.0	21.2	66.0
83423	01/31/2016	484.8	29.8	21.0	80.2

Source: INMET (Goiânia Station - OMM: 83423), 2017.

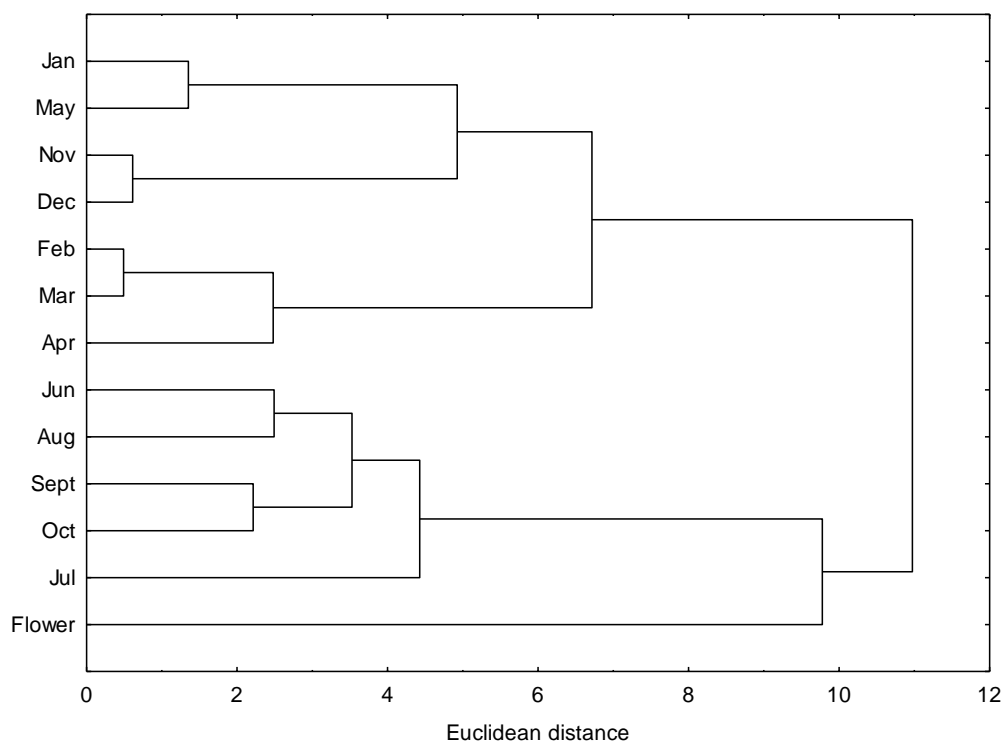
Figure 1- Dendrogram representing the similarity relations of the chemical composition of *C. adamantium* oils according to the method of Ward Minimization of variance. For this analysis were considered cis-eudesma-6,11-diene, γ -elemene, linalool, tricyclene, italicene epoxyde, α -guaiene, β -funebrene.




Figure 2- Scatterplot of PCA of the essential oils from the leaves of *C. adamantium* samples collected from Bela Vista/GO belonging to the clusters (I, II). ^aAxes referring to the scores of samples. ^b Axes referring to scores of volatile chemicals whose discriminant constituents are represented by vectors.

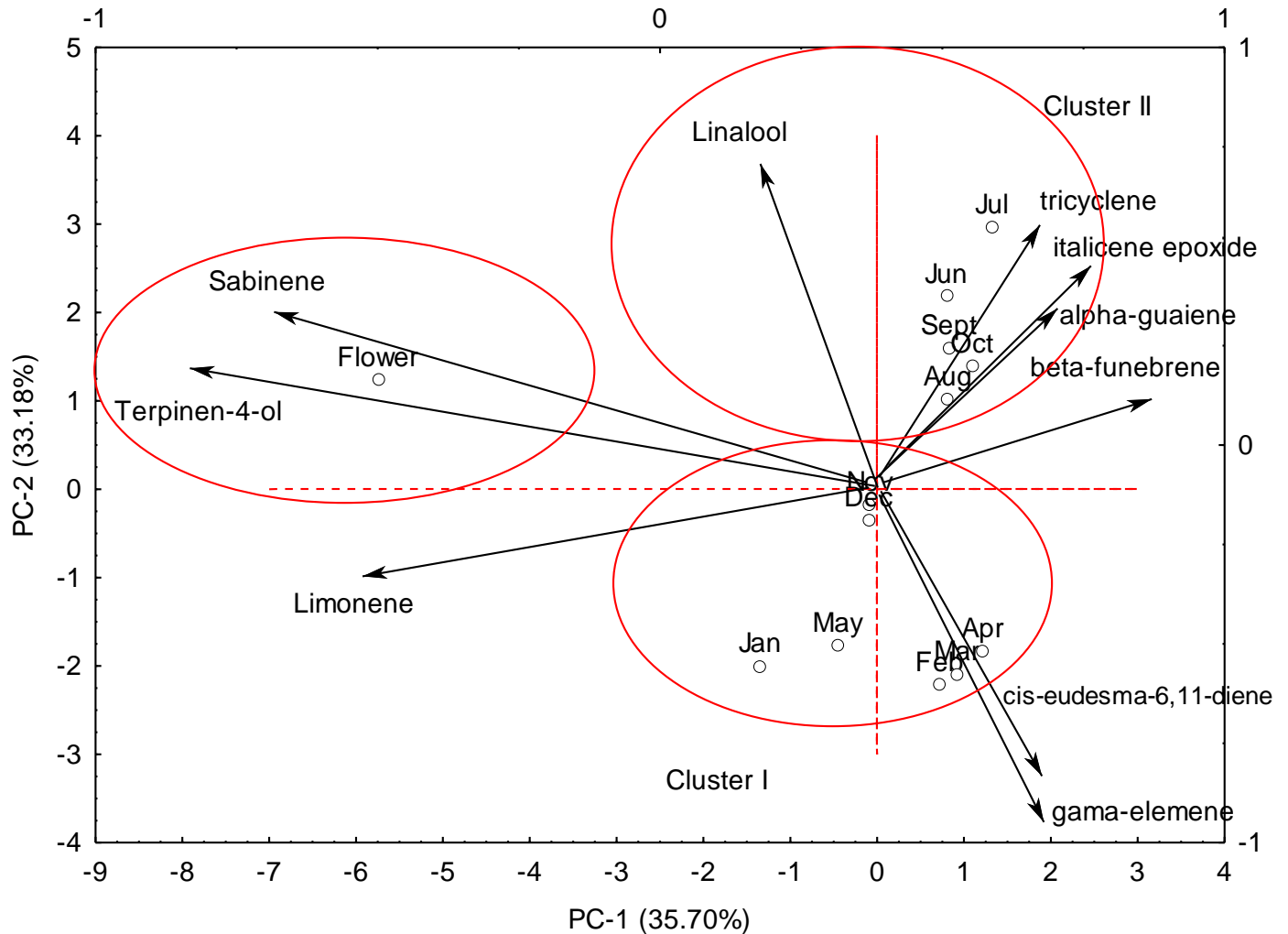



Table 3 Canonical discriminant analysis of *Campomanesia adamantium*.

Canonical discriminant				
	Eigenvalues functions	Canonical R	Wilk's Lambda	p-level
F1	5.99	0.93	0.1429	0.0015
Standardized Coefficients (for Canonical Variables)				
Tricyclene	0.6918			
Gama-elemene	-0.8140		0.2397	0.03
Eigenvalues	5.99		0.3241	0.008
Cumulative Proporcion	1			
Percent Correct				
		Cluster I	Cluster II	
		p=0.58	p=0.42	
Cluster I	100%	7	0	
Cluster II	100%	0	5	
Total	100%	7	5	

Table 4 – Content of phenols and tannins of *Campomanesia adamantium* leaves.

	Sample	Total phenols (%)	Total tannins (%)
Cluster I	Feb	4.28	2.91
Cluster I	Mar	5.08	3.09
Cluster I	Apr	5.60	2.85
Cluster I	May	5.39	3.14
Cluster II	Jun	5.75	2.82
Cluster II	Jul	6.38	3.68
Cluster II	Aug	7.84	4.08
Cluster II	Sept	9.56	4.84
Cluster II	Oct	7.21	3.27
Cluster I	Nov	4.25	3.17
Cluster I	Dec	3.75	2.25
Cluster I	Jan	6.64	3.31



Figure 3 – Seasonal variation of total phenols (TP) and total tannin (PP) from *Campomanesia adamantium* leaves (February 2015 to January 2016).

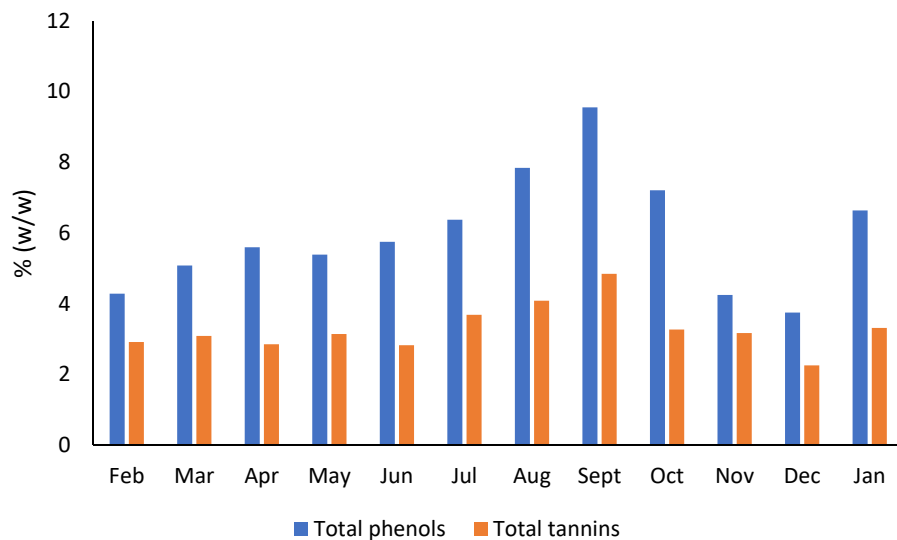


Table 2. Percentage of the chemical constituents of the volatile oils from *C. adamantium* leaves and flower collected in Bela Vista, Goiás.

Constituents	KI	Leaves-2015												Leaves-2016	Flower-2015
		Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan	Flower-Oct	
Tricyclene	926					10.2	3.0	3.8	3.33	8.6			0.6	9.1	
α -Pinene	939					20.6			5.43	12.8	18.7	16.4	2.4		
Verbenene	967					0.9	0.6	0.9	4.29	3.1					
Sabinene	975			0.5					8.32	5.1	7.9	7.6	3.8	20.5	
β -Pinene	979								1.2	0.5	0.2				
α -Terpinene	1017	0.9			1.3	0.6	0.5	1.0	0.37	0.5	0.6	0.6		4.9	
Limonene	1029	4.83	1.52	2.19	20.7	5.18	6.19	12.6	5.32	10.0	12.3	13.1	27.7	19.33	
Linalool	1096	0.9	0.7	0.4	1.1	6.6	8.7	3.8	3.81	1.3	1.6	1.6	0.6	10.0	
α -Terpineol	1188	0.8	0.8	0.6	1.0	1.7	4.0	3.1	1.63	1.2	1.4	1.6	1.1	4.58	
Methyl salicylate	1191													8.7	
neo-Dihydro carveol	1194	1.0	1.1	1.2	1.4	1.3	0.9	1.7	0.50	1.3	1.7	2.0	0.9		
α -Ylangene	1375					0.9	1.4	0.7	1.31	0.9	0.6	0.6			
β -Funebrene	1414	8.7	8.7	12.7	6.2	10.3	11.6	12.5	11.89	12.9	12.4	11.5	5.5	3.13	
α -Guaiene	1439	0.9	0.93	0.8	1.8	1.9	2.4	2.3	4.92	4.0	3.1	2.7	1.0		



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Table 2. Percentage of the chemical constituents of the volatile oils from *C. adamantium* leaves and flower collected in Bela Vista, Goiás.

Constituents	KI	Leaves-2015												Leaves-2016	Flower-2015
		Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan	Flower-Ooct	
γ-Elemene	1436	23.46	22.9	30.41	23.03	8.42	6.26	10.9	3.8	8.3	14.6	16.9	20.7		
Aromadendrene	1441	0.9	1.0	1.5	1.2	1.4	1.8	1.7	2.10	1.7	1.6	1.5	0.6	1.93	
allo-Aromadendrene	1460	1.9	1.4	0.8		0.5	0.7	0.5	0.93	0.4	0.2		1.7		
Dauca-5,8-diene	1472	3.5	6.4	3.9	3.7	3.8	1.3	3.4	1.94	0.3	1.3	1.9	4.4		
Bicyclogermacrene	1500	4.2	8.9	7.1	8.9	0.5	0.7	0.6	0.8	0.8	1.0	1.5	5.6	1.05	
cis-Eudesma-6,11-diene	1489	8.0	7.9	4.3	4.7	1.1	0.6	1.7	4.12	2.7	2.3	2.6	5.0		
β-Selinene	1490					0.4	0.4	0.4	0.72	0.3					
Epizonarene	1501			0.4		0.7	0.8	0.4	0.82	0.6	0.4	0.4			
trans-β-Guaiene	1502	0.6	0.6	0.8	0.8	0.6	1.5	-	0.41	0.4	0.7	0.8	0.6		
α-dehydro-ar-Himachalene	1517		0.5	0.8	0.4	0.3	-	0.4	0.65	0.3	0.3	0.3			
Italicene epoxide	1548	4.3	5.4	5.1	2.6	5.8	18.6	8.4	7.05	6.3	1.5	3.3	0.9		
trans-Dauca-4(11).7-diene	1557	3.3	4.1	4.5	2.9	2.2	2.0	4.0	3.38	1.7	1.8	1.8	1.7		
Maaliol	1567	1.5	1.6	3.5	1.0	1.0	0.2	2.0	2.39	1.3	1.6	1.6	0.7	3.84	
Thujopsan-2-α-ol	1587	0.7	0.8		0.5	0.4	1.3	0.7	0.91	0.6			0.4		
Viridiflorol	1592													2.21	


Table 2. Percentage of the chemical constituents of the volatile oils from *C. adamantium* leaves and flower collected in Bela Vista, Goiás.

Constituents	KI	Leaves-2015												Leaves-2016	Flower-2015	
		Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan	Flower-Ooct		
Cubeban-11-ol	1595															0.74
Rosifoliol	1600					0.7	1.2	1.0	0.50							1.10
Humulene epoxide II	1608		0.8	0.88					0.42	0.4		0.5				0.71
Muurolol	1642	0.5	0.6			0.6	0.5	0.7	0.81		0.4	0.4				
Cubenol	1646															0.83
Monoterpene hydrocarbons		5.7	1.5	2.7	22	37.4	10.3	18.3	24.9	40.6	39.7	37.7	34.5			53.83
Oxygenated monoterpenes		2.7	2.6	2.2	3.5	9.6	13.6	8.6	5.9	3.8	4.7	5.2	2.6			14.6
Sesquiterpene hydrocarbons		55.5	63.3	68	53.9	33.0	31.5	39.5	37.8	35.3	40.3	42.5	46.9			6.1
Oxygenated sesquiterpenes		7.0	9.2	10.0	4.1	9.0	23.2	14.5	12.7	8.6	3.5	6.1	2.0			9.4
Others		0.7	-	-	-	-	-	-	-	-	-	-	1.4			8.7
Total identified (%)		71.6	76.6	82.9	83.5	89.0	78.6	80.9	81.3	88.3	88.2	91.5	87.4			92.6



Table 2. Percentage of the chemical constituents of the volatile oils from *C. adamantium* leaves and flower collected in Bela Vista, Goiás.

Constituents	KI	Leaves-2015												Leaves- 2016	Flower- 2015
		Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec	Jan	Flower- Ooct	
Yield (%)		0.7	0.8	0.6	0.6	0.6	0.6	0.7	2.0	2.5	0.9	0.6	0.5	1.5	