# Variability in the Chemical Composition of *Eugenia biflora* Essential Oils from the Brazilian Amazon

Natural Product Communications Volume 15(12): 1–6 © The Author(s) 2019 Article reuse guidelines: sagepub.com/journals-permissions DOI: 10.1177/1934578X19892439 journals.sagepub.com/home/npx



Pablo Luis B. Figueiredo<sup>1,2</sup>, Henryck A. Fernandes<sup>3</sup>, Alberto Ray C. da Silva<sup>4</sup>, Nayara Sabrina F. Alves<sup>5</sup>, William N. Setzer<sup>6,7</sup>, Joyce Kelly R. da Silva<sup>1,5</sup>, and José Guilherme S. Maia<sup>1,8</sup>

## Abstract

*Eugenia* species are well known for their great economic potential as edible fruits. The leaves of 4 *Eugenia biflora* specimens (Ebi-1 to Ebi-4) were sampled in the Caratateua Island, Pará state, Brazilian Amazon. Then, the essential oils were hydrodistilled, analyzed by gas chromatography (GC) and GC mass spectroscopy, and their volatile compositions submitted to multivariate analysis (principal component analysis and hierarchical cluster analysis). Ebi-1 and Ebi-2 specimens were classified in the caryophyllene group, with significant content for the sesquiterpenes (*E*)-caryophyllene (16.8% and 11.4%) and caryophyllene oxide (28.6% and 20.5%). Ebi-3 specimen was grouped into the cadinane group, characterized by the presence of  $\alpha$ -cadinol (14.7%), an oxygenated sesquiterpene. EBI-4 specimen was inserted into the aromadendrane group, with the predominance of the sesquiterpenes globulol (9.8%), germacrene B (7.9%), and  $\gamma$ -elemene (3.1%). Based on the results, a remarkable chemical variability was observed in the oils of *Eugenia biflora* with occurrence in Pará state, North Brazil. This work is presenting for the first time its caryophyllene, cadinane, and aromadendrane profiles.

#### Keywords

Myrtaceae, sesquiterpene compounds, essential oil, chemical composition, multivariate analysis

Received: August 27th, 2019; Accepted: October 23rd, 2019.

Myrtaceae consists of 144 genera and 6019 species distributed mainly in the Southern Hemisphere with particular emphasis in Australia and South America.<sup>1,2</sup> Among the genera that belong to this family, *Eugenia* has one of the most significant numbers of species, which corresponds to approximately 1000 species.<sup>3</sup>

Species of the genus *Eugenia* are well known for their great economic potential evidenced by the commercial exploitation of edible fruits, woods, and essential oils and by their use as ornamental plants. Besides this, many taxa are employed in the traditional medicine of tropical and subtropical regions around the world.<sup>4-6</sup>

Essential oils produced by some species of *Eugenia* from the Amazon have critical pharmacological applications because of their biological activities. For instance, *E. uniflora* has antioxidant, antimicrobial, and antitumor potential,<sup>7-9</sup> and *E. egensis*, *E. flavescens*, *E. patrisii*, and *E. polystachya* have cytotoxic effects.<sup>10</sup>

The species *Eugenia biflora* (L.) DC. (syn. *E. acuminatissima* Miq., *E. alfaroana* Standl., *E. amanuensis* Steyerm.), popularly known as "Vassourinha," "Murta," or "Pedra-hume-caá",<sup>11-13</sup> is distributed in North, Central, and South America. The plant is native and not endemic to Brazil, occurring in Northern (Acre, Amazonas, Amapá, Pará, Rondônia, Roraima, and Tocantins states), Northeastern (Maranhão, Piauí states), and Midwestern (Goiás, Mato Grosso do Sul, Mato Grosso states) regions.<sup>13</sup>

<sup>2</sup>Departamento de Ciências Naturais, Centro de Ciências Sociais e Educação, Universidade do Estado do Pará, Belém, Brazil

<sup>3</sup>Programa Institucional de Bolsas de Iniciação Científica, Universidade Federal do Pará, Belém, Brazil

<sup>5</sup>Programa de Pós-Graduação em Biotecnologia, Universidade Federal do Pará, Belém, Brazil

<sup>6</sup>Department of Chemistry, University of Alabama in Huntsville, USA

Aromatic Plant Research Center, Lehi, UT, USA

<sup>8</sup>Programa de Pós-Graduação em Química, Universidade Federal do Maranhão, São Luís, Brazil

#### **Corresponding Author:**

Pablo Luis B. Figueiredo, Programa de Pós-Graduação em Química, Universidade Federal do Pará, Belém, PA 66075-110, Brazil. Email: pablolbf@ufpa.br



Creative Commons Non Commercial CC BY-NC: This article is distributed under the terms of the Creative Commons Attribution-NonCommercial 4.0 License (http://www.creativecommons.org/licenses/by-nc/4.0/) which permits non-commercial use, reproduction and distribution of the work without further permission provided the original work is attributed as specified on the SAGE and Open Access pages (https://us.sagepub.com/en-us/nam/open-access-at-sage).

<sup>&</sup>lt;sup>1</sup>Programa de Pós-Graduação em Química, Universidade Federal do Pará, Belém, Brazil

<sup>&</sup>lt;sup>4</sup>Faculdade Integrada Brasil Amazônia, Belém, Brazil

*E. biflora* is a shrub that can grow to a height of 0.5-3.0 m. The species has elliptical, oval-lanceolate-shaped leaves with flat margins and grooved midribs on the adaxial surface, while its inflorescences are raceme type, solitary, or overlapping, axillary or terminal, with rachis of 4-27 mm. The flowers have a persistent nonshowy bract, and its fruits present a globular, ellipsoid shape with a smooth surface.<sup>13</sup> The plant is employed by traditional Amazonian communities to treat diarrhea, thrush, intestinal inflammation, and hemorrhages.<sup>12,14</sup> The light petroleum extracts from leaves were rich in  $\beta$ -amyrin and eucalyptin,<sup>11</sup> showing antimicrobial activity, which also emphasizes its pharmacological potential.<sup>12</sup>

There is only 1 study of leaf essential oils of *E. biflora* previously reported, which was collected in the municipality of Maracanã, State of Pará, showing a high content of  $\alpha$ - and  $\beta$ -pinene and (*E*)-caryophyllene.<sup>15</sup> Based on the species potential, the present study aimed to investigate the variability of the chemical composition of different specimens that occur in the Brazilian Amazon, to contribute to the knowledge of its chemotaxonomy.

*E. biflora* belongs to the group of plants known in the Amazon as "Pedra-hume-caá" (pedra-hume means alumen and caá means leaf in Tupi-Guarani language). The group comprises, in addition to *E. biflora*, the species *Myrcia multiflora* and *Myrcia sylvatica*. These 3 species have a broad pharmacological application and generate income in traditional communities because of their cultivation and commercialization.<sup>12</sup>

Four specimens of *E. biflora* were collected and showed distinct chemical composition for their essential oils (Table 1). These oils were light yellow with a woody odor and presented yields of 0.4% to Ebi-1, 0.3% to Ebi-2 and Ebi-4, and 0.2% to Ebi-3 samples (Table 1). The quantification and identification by gas chromatography (GC) and gas chromatography mass spectroscopy (GC-MS) presented 78 constituents in the oils of the specimens, representing an average of 92.1% of the total oils content (Table 2). In oils, oxygenated sesquiterpenes (48.0%-74.1%) and sesquiterpene hydrocarbons (19.0%-45.6%) predominated. Monoterpenes were not identified in the oils. The compounds caryophyllene oxide (0.5%-28.6%), (*E*)-caryophyllene (1.8%-16.8%),  $\alpha$ -cadinol (6.2%-14.7%), globulol (0.3%-9.8%), and epi- $\alpha$ -murrolol (4.0%-8.4%) were the main constituents.

To classify the oils, a multivariate analysis (principal component analysis, PCA, and hierarchical cluster analysis) was

Table 1. Identification Data for the Specimens of Eugenia biflora.

Samples	Herbarium number	Local coordinates
Ebi-1	NR	1° 14'55.97"S / 48°26'32.61"W
Ebi-2	MG-229216	1°14'55.47"S / 48°26'32.31"W
Ebi-3	MG-229219	1°14'26.79"S / 48°25'39.29"W
Ebi-4	MG-229220	1°14'26.25"S / 48°25'40.16"W

 ${\rm NR}={\rm not}$  yet registered (in process). Botanical identification was made by comparison with authentic samples.

applied using the chemical components (>3%) as variables (Figures 1 and 2). The PCA explained 92.7% of the total data variability. The component PC1 explained 56.0% and displayed positive correlations with the variables  $\alpha$ -muurolol, copaborneol,  $\alpha$ -cadinol, *epi*- $\alpha$ -cadinol, spathulenol, viridiflorol,  $\gamma$ -elemene, germacrene B, and globulol. The component PC2 explained 36.7% and showed a positive correlation with the compounds caryophyllene oxide, 1-*epi*-cubenol, (*E*)-nerolidol, *epi*- $\alpha$ -muurolol, copaborneol,  $\alpha$ -cadinol, *epi*- $\alpha$ -cadinol, and spathulenol.

The specimens Ebi-1 and Ebi-2 were grouped in the caryophyllene group due to its significant content of the sesquiterpenes, (*E*)-caryophyllene (16.8% and 11.4%), and caryophyllene oxide (28.6% and 20.5%). The specimen Ebi-3 was grouped into the cadinane group characterized by the oxygenated sesquiterpene  $\alpha$ -cadinol (14.7%). The specimen Ebi-4 was grouped into the aromadendrane group with the predominance of the sesquiterpenes globulol (9.8%), germacrene B (7.9%), spathulenol (4.5%) and viridiflorol (3.5%) (Figure 2).

The sesquiterpenes (*E*)-caryophyllene and caryophyllene oxide are found in a significant number of plant species. These compounds have a significant anticancer activity, which affects the growth and proliferation of numerous tumor lineages, with low antioxidant activity.<sup>18</sup> Two different chemical profiles of *E. biflora*, collected in Pará state, were described by Pereira and coworkers.<sup>15</sup> The first one was characterized by the presence of the sesquiterpene hydrocarbon (*E*)-caryophyllene (15.36%), however, with the absence of caryophyllene oxide. The second profile was characterized by the presence of the monoterpene hydrocarbons  $\alpha$ - and  $\beta$ -pinene, with contents of 27.34% and 27.85%, respectively.

Species of *Eugenia* from the Amazon have shown significant intraspecific chemical variation. Leaf essential oils from *E. protenta*, collected from Northeastern Pará State, were rich in sesquiterpene hydrocarbons, such as germacrene D,  $\beta$ -elemene,  $\delta$ -cadinene, or keto-phenolic ethers, as dimethylxanthoxylin.<sup>19</sup> Similarly, oils of *E. flavescens*, from different regions of Pará State, were rich in sesquiterpenes with the germacrane skeleton (germacrene D and bicyclogermacrene), or the bisabolane skeleton ( $\beta$ -, (*E*)-*iso*- $\gamma$ - and (*E*)- $\gamma$ -bisabolene)<sup>10,15</sup>.

Additionally, oils of *E. patrisii* were characterized by (2E,6Z)-farnesol (23.2%) and (2E,6E)-farnesol (34.5%), or by a mixture of sesquiterpene hydrocarbons characterized by the presence of *trans*-cadina-1,4-diene (16.5%), *trans*-muurola-3,5-diene (13.3%), (*E*)-caryophyllene (11.1%), and  $\alpha$ -cubebene (9.8%), as its main compounds.<sup>10,15</sup>

# Conclusion

At least 5 *E. biflora* profiles are occurring in the State of Pará, at Brazilian Amazon. In the oils of these profiles oxygenated sesquiterpenes and sesquiterpene hydrocarbons predominated.

IR <sub>(C)</sub>	IR <sub>(L)</sub>	Constituents	Ebi-1	Ebi-2	Ebi-3	Ebi-4
858	857 <sup>b</sup>	Ethyl-benzene	0.3	0.1		
904	907 <sup>a</sup>	Butyl propanoate	0.9	0.2	0.1	0.1
999	1000 <sup>a</sup>	<i>n</i> -Decane	0.2			
1338	1335 <sup>a</sup>	δ-Elemene	0.2		0.1	0.3
1352	1345 <sup>a</sup>	α-Cubebene		0.1	0.1	0.1
1373	1373 <sup>a</sup>	α-Ylangene			0.1	0.1
1377	1374 <sup>a</sup>	α-Copaene	1.7	3.1	1.0	0.9
1381	1378 <sup>a</sup>	(3Z)-Hexenyl hexenoate				0.1
1387	1387 <sup>a</sup>	β-Bourbonene		0.5	0.1	0.1
1391	1387 <sup>a</sup>	β-Cubebene			0.2	
1394	1389 <sup>a</sup>	β-Elemene	0.7	1.8	0.9	2.6
1411	1409 <sup>a</sup>	α-Gurjunene				0.1
1422	1417 <sup>a</sup>	(E)-Caryophyllene	16.8	11.4	1.8	9.8
1431	1430 <sup>a</sup>	β-Copaene	0.4	0.4	0.7	0.5
1433	1432 <sup>b</sup>	γ-Elemene		0.5	0.3	3.1
1436	1432 <sup>b</sup>	trans-a-Bergamotene		0.2		
1440	1437 <sup>a</sup>	α-Guaiene			0.4	0.8
1442	1439 <sup>a</sup>	Aromadendrene		0.2		
1445	1442 <sup>a</sup>	6,9-Guaiadiene			0.3	0.4
1451	1447 <sup>b</sup>	Isogermacrene D			0.1	0.1
1454	1452 <sup>a</sup>	α-Humulene	1.6	1.0	0.4	1.1
1462	1458 <sup>a</sup>	allo-Aromadendrene			0.2	
1462	1464 <sup>a</sup>	9-epi-(E)-Caryophyllene		0.2		0.5
1477	1478 <sup>a</sup>	γ-Muurolene	0.7	0.8	1.6	1.1
1481	1483 <sup>a</sup>	α-Amorphene	0.4			
1484	1476 <sup>b</sup>	Selina-4,11-diene <sup>c</sup>				0.1
1484	1484 <sup>a</sup>	Germacrene D		0.7	1.7	2.2
1487	1492 <sup>a</sup>	β-Selinene		0.4	0.6	1.0
1494	1493 <sup>a</sup>	trans-Muurola-4(14),5-diene				0.2
1494	1493 <sup>a</sup>	epi-Cubebol	1.8			
1495	1496 <sup>a</sup>	Valencene		0.9	1.4	
1496	1496 <sup>a</sup>	Viridiflorene				2.1
1498	1500 <sup>a</sup>	Bicyclogermacrene			0.2	2.1
1501	$1500^{a}$	α-Muurolene	0.7	0.6	1.4	0.8
1506	1505 <sup>b</sup>	α-Bulnesene			0.1	0.2
1509	$1508^{\mathrm{b}}$	β-Bisabolene			0.1	0.2
1515	1513 <sup>a</sup>	y-Cadinene	2.9	0.8	2.2	0.9
1519	1520 <sup>a</sup>	7 <i>-epi</i> -α-Selinene			0.4	0.7
1524	1522 <sup>a</sup>	δ-Cadinene				2.1
1527	1521 <sup>a</sup>	trans-Calamenene	2.1	1.5	1.5	
1524	1522 <sup>a</sup>	δ-Cadinene				2.1
1536	$1540^{b}$	Selina-4(15),7(11)-diene				0.5
1538	1532 <sup>a</sup>	γ-Cuprenene		1.5		
1540	1537 <sup>a</sup>	α-Cadinene			0.4	0.4
1542	1545 <sup>a</sup>	Selina-3,7(11)-diene				0.5
1549	1548 <sup>a</sup>	Elemol		0.3	1.8	0.7
1558	1559 <sup>a</sup>	Germacrene B		1.1	0.7	7.9
1564	1561 <sup>a</sup>	(E)-Nerolidol	1.4	3.6	1.6	0.5
1569	1567 <sup>a</sup>	Palustrol				1.4
1579	1577 <sup>a</sup>	Spathulenol	2.2	2.4	4.9	4.5
1583	1584 <sup>a</sup>	Caryophyllene oxide	28.6	20.5	7.5	0.5
1588	1590 <sup>a</sup>	Globulol		0.3		9.8

Table 2. Essential Oil Yield and Composition of 4 Samples of Eugenia biflora.

(Continued)

IR <sub>(C)</sub>	IR <sub>(L)</sub>	Constituents	Ebi-1	Ebi-2	Ebi-3	Ebi-4
1592	1592 <sup>a</sup>	Viridiflorol	2.0	1.6	2.9	3.5
1594	1595 <sup>a</sup>	Cubeban-11-ol	0.4		1.8	1.4
1602	1600 <sup>a</sup>	Rosifoliol				1.5
1605	1602 <sup>a</sup>	Ledol	1.0			
1605	1613 <sup>a</sup>	Copaborneol			5.6	
1609	$1608^{a}$	Humulene epoxide II	1.1	1.1	0.4	0.4
1615	1618 <sup>a</sup>	1,10-di-epi-Cubenol			0.9	0.3
1620	1618 <sup>a</sup>	Junenol			2.0	1.3
1626	1624 <sup>b</sup>	epi-y-Eudesmol		1.8	2.2	
1629	1627 <sup>a</sup>	1- <i>epi</i> -Cubenol	6.4	6.6	3.3	1.6
1633	1630 <sup>a</sup>	γ-Eudesmol			1.8	0.8
1637	1639 <sup>a</sup>	Caryophylla-4(12),8(13)-dien-5β-ol		2.6		
1641	1638 <sup>a</sup>	<i>epi</i> -α-Cadinol (τ-cadinol)			4.9	1.9
1642	1640 <sup>a</sup>	epi-a-Murrolol (7-muurolol)	8.4	7.6	8.3	4.0
1647	1644 <sup>a</sup>	α-Muurolol (Torreyol)	2.4	3.0	4.8	2.1
1651	1649 <sup>a</sup>	β-Eudesmol			1.6	0.7
1653	1652 <sup>a</sup>	Himachalol				0.9
1655	1652 <sup>a</sup>	α-Cadinol	6.2	6.3	14.7	7.5
1657	1661 <sup>a</sup>	Allohimachalol		0.9		
1659	1660 <sup>a</sup>	cis-Calamenen-10-ol			0.5	
1666	1665 <sup>a</sup>	Intermedeol				0.5
1669	1668 <sup>a</sup>	trans-Calamenen-10-ol	0.3	0.2	0.2	
1671	1668 <sup>a</sup>	14-Hydroxy-9-epi-(E)-caryophyllene		1.1	0.8	0.5
1686	1685 <sup>a</sup>	Germacra-4(15),5,10(14)-trien-1-α-ol		1.4	1.0	0.3
1696	1696 <sup>b</sup>	Juniper camphor		0.4	0.5	1.4
1701	1702 <sup>a</sup>	10-nor-Calamenen-10-one			0.1	
quiterpene hydrocarbons		28.2	27.7	19.0	45.6	
xygenated sesquiterpenes		62.2	61.7	74.1	48.0	
thers			1.4	0.3	0.1	0.2
<b>'otal (%</b> )			91.8	89.7	93.2	93.8
il yield (%)			0.4	0.3	0.2	0.3

Table 2. Continued

 $RI_{(C)}$ : calculated retention time;  $RI_{(L)}$ : literature retention time; <sup>16,17</sup>; Bold: main constituents (above 5%).

<sup>a</sup>Adams (2007).<sup>17</sup>

<sup>b</sup>Mondello (2011).<sup>16</sup>

<sup>c</sup>correct stereoisomer was not determined.

This work reports the caryophyllene, cadinane, and aromadendrane profiles for the first time.

# Materials and Methods

# Plant Material

Leaves of 4 specimens of *E. biflora* were collected in Caratateua Island, State of Pará, Brazil. Plant vouchers were identified by Professor Marcos Sobral, a Brazilian Myrtaceae specialist, and deposited in the Herbarium of the Museu Emilio Goeldi, city of Belém, State of Pará, Brazil. The coordinates for each collected specimen and the voucher number are listed in Table 1. About 150 g of leaves from each plant were collected and dried for 2 days at room temperature. Dried leaves (60 g) were then subjected to hydrodistillation.

# Extraction and Oil Composition Analysis

Essential oils were hydrodistilled using a Clevenger-type apparatus (3 hours), according to Gottlieb and Magalhães.<sup>20</sup> Then, the oils were dried over anhydrous sodium sulfate, and the total oil yields are expressed as mL/100 g of dried material. The oil composition analysis was performed by GC-MS, using a Shimadzu instrument Model QP 2010 ultra (Shimadzu, Tokyo, Japan) equipped with an Rtx-5MS (30 m × 0.25 mm; 0.25 µm film thickness) silica capillary column (Restek, Bellefonte, PA, USA). Helium was used as the carrier gas, with a linear velocity of 1.0 mL/min at 57.5 kPa; split injection (split ratio 1:20) of 1 µL of a hexane solution (5 µL oil: 500 µL hexane); injector and interface temperatures were 250°C. The programmed temperature was 60 to 240°C (3°C/min), followed by an isotherm of 10 minutes. Electron impact mass spectrometry: electron energy was 70 eV,

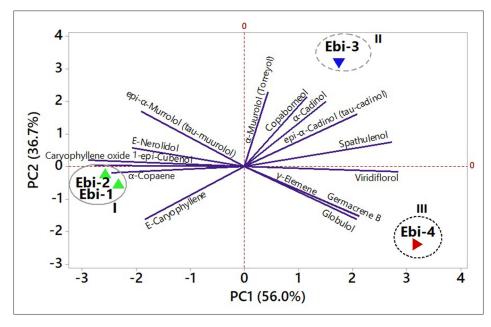


Figure 1. Biplot (principal component analysis) resulting from the analysis of the oils of Eugenia biflora.

and the ion source temperature was 200°C. The mass spectra were obtained by automatic scanning at every 0.3 second, with mass fragments in the range of 35-400 m/z. The oil constituents were identified by comparison of their mass spectra and retention indices with those available in commercial libraries, such as Adams<sup>16</sup> and FFNSC 2/Mondello.<sup>17</sup> Retention indices were calculated using *n*-alkane standard solutions (C8-C40, Sigma-Aldrich, St. Louis, MO, USA) in the same chromatographic conditions.<sup>21</sup> GC flame ionization detector (FID) analysis was

carried out on a Shimadzu QP-2010 instrument equipped with an FID detector in the same conditions, except that hydrogen was used as the carrier gas. The oil constituents percentual was computed from GC-FID peak areas.

# Statistical Analysis

The multivariate analysis was performed by using as variables the constituents of oils with content above 3.0%. The data

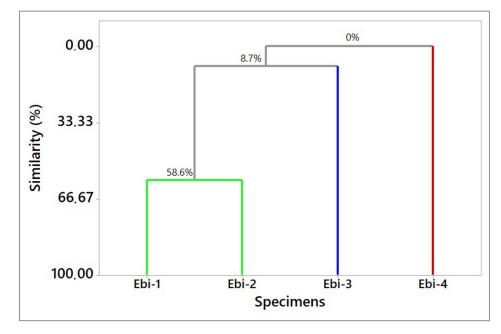


Figure 2. Dendrogram representing the similarity relation for the oils of Eugenia biflora.

matrix was standardized by subtracting the mean and then dividing it by the SD. For hierarchical cluster analysis, the complete linkage method and the Euclidean distance were used. All analyses were performed using the software Minitab (free 390 version, Minitab Inc., State College, PA, USA).

#### Acknowledgments

We are grateful to CNPq and Capes, Brazilian government funding agencies, for scientific research support.

## **Declaration of Conflicting Interests**

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

## Funding

The author(s) received no financial support for the research, authorship, and/or publication of this article.

# **ORCID ID**

Pablo Luis B. Figueiredo D https://orcid.org/0000-0003-1365-3513 William N. Setzer D https://orcid.org/0000-0002-3639-0528

## References

- Wilson PG. Myrtaceae. In: Kubitzki K, ed. The Families and Genera of Vascular Plants: Flowering Plants Eudicots Sapindales, Cucurbitales, Myrtaceae. 1st ed. Nova Deli: Springer-Verlag Berlin Heidelberg; 2011:436.
- WSCP. World Checklist of Selected Plant Families. Facilitated by the Royal Botanic Gardens. Kew; 2012.
- Van der Merwe MM, Van Wyk AE, Botha AM. Molecular phylogenetic analysis of *Eugenia* L. (Myrtaceae), with emphasis on southern African taxa. *Plant Syst Evol.* 2005;251(1):21-34.
- Queiroz JMG, Suzuki MCM, Motta APR, Nogueira JMR, de CEM. Aspectos populares e científicos do uso de espécies de *Eugenia* como fitoterápico. *Rev Fitos.* 2015;9(2):87-100.
- Donadio LC, Moro FV. Potential of Brazilian eugenia myrtaceaeas ornamental and as a fruit crop. In: *Acta Horticulturae*. Leuven, Belgium: International Society for Horticultural Science (ISHS); 2004:65-68.
- Mitra SK, Irenaeus TKS, Gurung MR, Pathak PK. Taxonomy and importance of myrtaceae. In: *Acta Horticulturae*. Leuven, Belgium: International Society for Horticultural Science (ISHS); 2012:23-34.
- Dos Santos JFS, Rocha JE, Bezerra CF, et al. Chemical composition, antifungal activity and potential anti-virulence evaluation

of the *Eugenia uniflora* essential oil against *Candida* spp. *Food Chem*. 2018;261:233-239.

- Victoria FN, Lenardão EJ, Savegnago L, et al. Essential oil of the leaves of *Eugenia uniflora* L.: antioxidant and antimicrobial properties. *Food Chem Toxicol.* 2012;50(8):2668-2674.
- Figueiredo PLB, Pinto LC, da Costa JS, et al. Composition, antioxidant capacity and cytotoxic activity of *Eugenia uniflora* L. chemotype-oils from the Amazon. J Ethnopharmacol. 2019;232:30-38.
- da Silva J, Andrade E, Barreto L, et al. Chemical composition of four essential oils of *Eugenia* from the brazilian amazon and their cytotoxic and antioxidant activity. *Medicines*. 2017;4(3):51.
- Gottlieb OR, da Silva ML, Maia JGS. Eucalyptin from *Eugenia* and *Myrcia* species. *Phytochemistry*. 1972;11(3):1185.
- 12. da Silva FKS, do Rosário AS, Secco RS, Zoghbi MGB. A survey of the species known as pedra-ume-caá (Myrtaceae) with emphasis with emphasis on those species recognized and commercialized in the city of Belém, Pará, Brazil. *Biota Amaz.* 2015;5(1):7-15.
- Sobral M, Proença C, Souza M, Mazine F, Lucas E. Myrtaceae in lista de espécies da flora do Brasil. *Jard Bot Rio de Janeiro*. 2019. Available in: http://floradobrasil.jbrj.gov.br
- Tropicos.org. *Eugenia biflora* (L.) DC. Missouri Botanical. https:// www.tropicos.org/Name/22101722. Accessed October 2019.
- Pereira RA, Zoghbi MdasGB, Bastos MdeNdoC. Essential oils of twelve species of Myrtaceae growing wild in the sandbank of the Resex Maracanã, state of Pará, Brazil. *Journal of Essential Oil Bearing Plants*. 2010;13(4):440-450.
- Mondello L. FFNSC 2: Flavors and Fragrances of Natural and Synthetic Compounds, Mass Spectral Database. New york: John Wiley & Sons Inc; 2011.
- Adams RP. Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry. Carol Stream, Ilinois, USA: Allured Publishing Corporation; 2007.
- Fidyt K, Fiedorowicz A, Strządała L, Szumny A. β-caryophyllene and β-caryophyllene oxide-natural compounds of anticancer and analgesic properties. *Cancer Med.* 2016;5(10):3007-3017.
- Zoghbi MGB, Guilhon GMSP, Sarges FN, Pereira RA, Oliveira J. Chemical variability of the volatiles from the leaves of *Eugenia* protenta McVaugh (Myrtaceae) growing wild in the North of Brazil. *Biochem Syst Ecol.* 2011;39(4-6):660-665.
- Gottlieb OR, Magalhães MT. Modified distillation trap. *Chemist* Analyst. 1960;49(4):114.
- van Den Dool H, Dec. Kratz P. A generalization of the retention index system including linear temperature programmed gas—liquid partition chromatography. J Chromatogr A. 1963;11:463-471.