Revista Brasileira de Farmacognosia
Brazilian Journal of Pharmacognosy
23(3): 433-440, May/Jun. 2013

Chemical variability in the essential oils from leaves of *Syzygium jambos*

Wilma P. Rezende,[1] Leonardo L. Borges,[*,1] Nilda M. Alves,[2] Pedro H. Ferri,[3] José R. Paula[1]

[1]Laboratório de Pesquisa de Produtos Naturais, Faculdade de Farmácia, Universidade Federal de Goiás, Brazil,
[2]Escola Superior de Ciências da Saúde, Departamento de Farmácia e Bioquímica, Universidade de Rio Verde, Brazil,
[3]Laboratório de Bioatividade Molecular, Instituto de Química, Universidade Federal de Goiás, Brazil.

Article

Received 20 Dec 2012
Accepted 15 Apr 2013
Available online 14 May 2013

Abstract: *Syzygium jambos* (L.) Alston, Myrtaceae, occurs in tropical regions and is a widespread medicinal plant used to treat several diseases, such as hemorrhage, dysentery, diabetes, inflammation, diabetes and gastrointestinal disorders. Leaf essential oils of ten specimens of *S. jambos* collected from two localities of Central Brazilian Cerrado were investigated by GC-MS. Soil and foliar nutrients were analyzed to determine the mineral compositions. The aims of this study was to evaluate the influence of environmental factors on chemical composition of leaf essential oils of *S. jambos*. Studies on the influence of environmental factors over composition of essential oils are important because they contribute data for its cultivation, harvest and establish parameters to essential oil components. The data were analyzed using *stepwise* Multiple Regression and Cluster Analysis, and the results suggest that the main factor capable to influence the chemical composition of leaf essential oils was the collection period and the collection site had a minor effect. The results also suggest that the leaf essential oils composition was influenced mainly by foliar nutrients (N, Mn, Co, Fe, S and Mg) and soil nutrients (Na, Al, S and H+Al). The compound with the best model obtained was the (*E*)-caryophyllene, with a coefficient of determination equal 0.8113.

Keywords:
medicinal plants
Myrtaceae
seasonality
mineral nutrients
statistical analysis

ISSN 0102-695X
DOI: 10.1590/S0102-695X2013005000035

Introduction

The Brazilian Cerrado stands out as the richest source of biodiversity, with about 6500 plant species cataloged and approximately 220 of which have medicinal uses (MMA, 2009). Several studies by authors suggest that, the percentage of species present in this biome may represent 20 to 50% of the total found in the country (Machado et al., 2004). The Myrtaceae family is one of the principal floras, with 23 genera and about 130 species, and many species are used popularly against gastrointestinal disorders, infectious diseases and hemorrhagic conditions. This family also includes many species that are characterized by the presence of essential oils (Rodrigues & Carvalho, 2001; Holetz et al., 2002; Amaral et al., 2006; Gondim et al., 2006; Sá et al., 2012). Among the species of this family, *Syzygium jambos* (L.) Alston (syn *Eugenia jambos*) stands out due its medicinal properties.

This genus has been reported for their different medicinal uses. *Syzygium cumini, S. aromaticum, Syzygium jambolanum* and *Syzygium jambos* are the most pharmacologically studied species, and have been recommended to treat haemorrhage, dysentery and gastrointestinal disorders (Moreira, 1978; Cruz, 1979), diabetes (Kelkar & Kaklij, 1996; Stanely Mainzen et al., 1998) and inflammation (Kim et al., 1998; Muruganandan et al., 2001). They have also been employed as sedative and anticonvulsivant (De Lima et al., 1998), as antihypertensive (Bhargava et al., 1968), against herpes virus (Kurokawa et al., 1998) and as inhibitor of histamine release ((Kim et al., 1998). Djipa et al., (2000) tested acetone and aqueous extracts from the barks of *S. jambos* for antimicrobial activity *in vitro* by the agar dilution method in petri dishes, and both extracts showed activity against *Staphylococcus aureus, Yersinia enterocolitica* and coagulase negative staphylococci among which *Staphylococcus hominis*, *Staphylococcus cohnii* and *Staphylococcus warneri.* The leaves of *S. jambos* are widely used in folk medicine to inflammation, digestive ulcers and high fever (Kan, 1987; Slowing et al., 1994a; 1994b; 1996; Rodrigues & Carvalho, 2001; Souza et al., 2002; Di Stasi et al., 2002; Pessini et al., 2003; Fiuza et al., 2008).

One property known of essential oils is their antimicrobial activity, particularly antibacterial and antifungal activities. Some of these species include the following: *Syzygium aromaticum* (L.) Merrirl et L. M. Perry, *Thymus* sp., *Lavandula* sp., *Origanum vulgare* L., *Rosmarinus officinalis* L. and *Eucalyptus globulus* Labill (Simões & Spitzer, 2004; Cunha, 2005).

It is important consider the chemical variations in essential oils caused by genetic, physiological or environmental variables when domesticating and improving plants of medicinal interest. Therefore, it is necessary studies that demonstrate that the chemical composition of the essential oils, although genetically determined, can suffer influence as a result of several environmental factors especially when referring to vegetal material used in chemical, pharmacological and agronomic studies that aim to obtain herbal medicines, because pharmacological properties can differ due to differences in essential oil composition (Lorenzi & Matos, 2002; Bergo et al., 2005; Lima et al., 2006; Potzernheim et al., 2006; Paula et al., 2011). The aim of our investigation was to study the chemical variability and environmental influence of the essential oil of *S. jambos* leaves in two samples collected from two different sites.

Materials and Methods

Plant material

Leaves of five specimens of *Syzygium jambos* (L.) Alston, Myrtaceae, were collected in the municipality of Rio Verde, Goiás state, Brazil (17º 48' 33,9" S; 50º 56' 39,1" W; 710 m), (17º 46' 33,6" S; 50º 54' 13,2" W; 688 m), (17º 46' 27,1" S; 50º 54' 52,2" W; 750 m), (17º 46' 09,6" S; 50º 54' 52,6" W; 781 m), (17º 46' 41,2" S; 50º 56' 43,1" W; 758 m) and five specimens in the municipality of Nova América, Goiás state, Brazil (15º 01' 12" S; 49º 52' 33,4" W; 782 m), (15º 01' 48,7" S; 49º 51' 27,9" W; 657 m), (15º 01' 48,6" S; 49º 51' 29,9" W; 652 m), (15º 02' 58,5" S; 49º 51' 53,4" W; 614 m). All samples were collected twice, in January and July 2011. The plants were identified by Prof. José Realino de Paula and a voucher was deposited at the Herbarium of the Federal University of Goiás under code number 47579. The leaf samples air-were dried in a chamber at 40 °C and ground into a powder.

Essential oil extraction

The essential oils from leaves of *S. jambos* were submitted to hydrodistillation in a modified Clevenger-type apparatus (2 h). Each essential oil was dried over anhydrous sodium sulfate and stored at - 20 °C for further analysis.

Essential oil analyses

Leaf essential oils obtained were analyzed using a gas chromatograph coupled to a mass selective detector (GC-MS), Shimadzu QP5050A, using an ionization voltage of 70 eV. A fused silica capillary column was utilized (CBP - 5; 30 m x 0.25 mm x 0.25 μm) and helium was used as the carrier gas at a flow rate of 1 mL min^{-1}. The temperature program used was as follows: ramp up from 60 to 240 °C at 3 °C min^{-1}, increase to 280 °C at 10 °C min^{-1}, and complete with 10 min at 280 °C. The injection volume was 1 μL diluted with CH_2Cl_2 at a ratio 1:5. The essential oil constituents were identified by comparing their mass spectra with those from the National Institute of Standards and Technology (NIST, 1998), as well as by comparing the mass spectra and calculated linear retention indices (RI) with values in the literature (Adams, 2007). Retention indices were obtained by co-injection with a mixture of linear hydrocarbons, C9-C22 (Sigma, USA) and calculated using the equation of Van Den Dool & Kratz (1963). The percentage of each component was calculated to normalize for the area in the chromatogram obtained using a Varian gas chromatograph (FID) equipped with a ZB-5 fused silica capillary column that was 30 m X 0.25 nm with 0.25 μm film thickness (5% phenylmethylpolysiloxane). The following temperature program was used: increase from 60 to 240 °C at 3 °C min^{-1}, followed by an increase to 280 °C at 10 °C min^{-1}, and complete with 10 min at 280 °C. The carrier gas was N_2, at a flow rate of 1.0 mL/min; the injector port and detector temperatures were 220 and 240 °C, respectively. Samples were injected by splitting, and the split ratio was 1:20.

Chemical analysis of leaves and soil

Chemical analysis of soil and leaf samples were performed at the Solocria Agricultural Laboratory, School of Agronomy, Federal University of Goiás, following standard procedures (Silva, 2009). For the analysis of foliar nutrients, the nitrogen (N) was extracted by digestion with H_2SO_4 and catalysts. The minerals phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S), copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) were extracted by digestion with $HClO_4$ and HNO_3. For the analysis of nutrients in the soil samples were collected at a depth of 0-20 cm in four locations around each specimen of *S. jambos*, subsequently homogenized and then air dried. A mass of 500 g was packed in plastic bags. The pH was determined in a volume of water-soil at 1:1. Ca, Mg and Al were extracted with KCl 1M, and P, K, Zn, Cu, Fe and Mn were extracted with Mehlich's solution. Organic matter (OM), cation exchange capacity (CEC), potential acidity (H+Al), base saturation (V) and aluminum saturation (m) were determined by standard methods (Silva, 2009). The quantitative determination of minerals in leaves and soil was performed according to the methodology described by (Silva, 2009). Nitrogen was determined by distillation (semi-micro Kjeldahl method), phosphorus by colorimetry, potassium by flame photometry and sulfur by turbidimetry. Calcium, magnesium, copper, iron, manganese and zinc were determined by atomic absorption.

Climate data

The monthly climate data (average temperature and average daily precipitation) for the collection months were collected from the official site of the National Institute for Space Research (Instituto Nacional de Pesquisas Espaciais).

Statistical analysis

The relationship between the components found in leaf essential oils (dependent variables) from *S. jambos* and the environmental factors (independent variables) were investigated by *stepwise* Multiple Regression and Pearson's Correlation Analysis implemented using SAS GLM and SAS CORR procedure, respectively (Draper & Smith, 1981). Cluster Analysis was also applied to the study of similarity of samples on the basis of constituent distribution and hierarchical clustering was performed according to Ward's variance minimizing method (Ward, 1963). Prior to the Cluster Analysis, the data were preprocessed by auto-scaling and mean centering. All procedures were performed using the software SAS (Statystical Analysis System) and Statistica 7 (Inc, 2002; Stat Soft, 2004).

Results and Discussion

The environmental variables are presented in Tables 1, 2, 3 and 4. The compounds of leaf essential oils analyzed are shown in Table 5.

Table 1. Climate data for the collection sites in the period of January 2011 and July 2011. Mean precipitation (mm) and mean temperature (ºC).

Sample	Precipitation (mm)	Temperature (ºC)
NA01/Jan/2011	11.93	24.48
NA02/Jan/2011	11.93	24.48
NA03/Jan/2011	11.93	24.48
NA04/Jan/2011	11.93	24.48
NA05/Jan/2011	11.93	24.48
RV01/Jan/2011	8.29	23.9
RV02/Jan/2011	8.29	23.9
RV03/Jan/2011	8.29	23.9
RV04/Jan/2011	8.29	23.9
RV05/Jan/2011	8.29	23.9
NA01/Jul/2011	-	21.62
NA02/Jul/2011	-	21.62
NA03/Jul/2011	-	21.62
NA04/Jul/2011	-	21.62
NA05/Jul/2011	-	21.62
RV01/Jul/2011	-	21.5
RV02/Jul/2011	-	21.5
RV03/Jul/2011	-	21.5
RV04/Jul/2011	-	21.5
RV05/Jul/2011	-	21.5

NA: Nova América; RV: Rio Verde.

Table 2. Levels of mineral nutrients and fertility parameters of soil from each sample collection site.

Sample	Cu mg/dm^3	Fe mg/dm^3	Mn mg/dm^3	Zn mg/dm^3	P mg/dm^3	K mg/dm^3	Ca mg/dm^3	Mg mg/dm^3
NA01/Jan/2011	6.00	310.00	34.00	20.00	1.00	10.00	5.00	1.70
NA02/Jan/2011	5.00	286.00	25.00	23.00	0.90	8.00	5.20	1.60
NA03/Jan/2011	5.00	276.00	24.00	22.00	1.20	10.60	4.90	1.50
NA04/Jan/2011	4.00	240.00	20.00	21.00	1.30	9.80	5.40	1.70
NA05/Jan/2011	7.00	243.00	18.00	19.00	1.20	8.40	4.70	2.20
RV01/Jan/2011	8.00	370.00	25.00	20.00	1.00	8.00	5.20	1.60
RV02/Jan/2011	5.00	305.00	22.00	21.00	1.00	8.00	5.50	1.60
RV03/Jan/2011	6.00	228.00	47.00	16.00	1.10	9.20	6.00	1.40
RV04/Jan/2011	6.00	202.00	61.00	16.00	1.40	9.60	6.20	1.60
RV05/Jan/2011	5.00	294.00	29.00	18.00	1.00	8.80	5.80	1.80
NA01/Jul/2011	4.00	241.00	29.00	11.00	1.00	7.60	5.20	1.70
NA02/Jul/2011	2.00	154.00	20.00	10.00	1.10	6.80	4.20	1.80
NA03/Jul/2011	3.00	367.00	19.00	11.00	1.00	8.00	4.40	1.60
NA04/Jul/2011	3.00	141.00	22.00	14.00	1.10	8.80	6.20	1.50
NA05/Jul/2011	3.00	148.00	24.00	14.00	1.20	8.60	6.50	1.60

RV01/Jul/2011	4.00	270.00	28.00	13.00	1.00	4.80	5.00	2.50
RV02/Jul/2011	4.00	363.00	22.00	14.00	1.10	6.40	6.00	1.80
RV03/Jul/2011	4.00	367.00	45.00	11.00	1.20	8.60	5.20	1.70
RV04/Jul/2011	6.00	345.00	58.00	11.00	1.20	8.00	5.40	1.60
RV05/Jul/2011	2.00	311.00	53.00	13.00	1.40	8.60	5.10	1.80

NA: Nova América; RV: Rio Verde.

Table 3. Levels of mineral nutrients and fertility parameters of soil from each collection site.

Sample	H+Al cmolc/dm³	Al cmolc/dm³	CEC cmolc/dm³	O.M. %	M %	V %	Ca/CEC %	Mg/CEC %	K/CEC %
NA01/Jan/2011	2.1	0.0	4.54	7.00	0.00	53.66	33.04	8.81	11.23
NA02/Jan/2011	2.3	0.0	6.70	14.00	0.00	65.63	43.28	16.42	5.67
NA03/Jan/2011	1.3	0.0	7.58	33.00	0.00	76.22	47.49	17.15	11.35
NA04/Jan/2011	1.8	0.0	11.07	67.00	0.00	88.27	73.17	9.03	5.87
NA05/Jan/2011	2.7	0.0	5.87	8.00	0.00	54.03	32.37	17.04	4.26
RV01/Jan/2011	4.0	0.1	6.82	8.00	3.44	41.39	26.39	13.20	1.61
RV02/Jan/2011	2.9	0.0	8.35	12.00	0.00	65.24	47.90	15.57	1.56
RV03/Jan/2011	1.9	0.0	8.26	11.00	0.00	76.95	61.74	10.90	4.00
RV04/Jan/2011	2.6	0.0	6.49	10.00	0.00	59.96	40.06	16.95	2.62
RV05/Jan/2011	2.8	0.0	9.30	14.00	0.00	69.85	50.54	13.98	5.05
NA01/Jul/2011	2.6	0.1	6.52	23.00	2.50	60.15	33.74	10.74	15.34
NA02/Jul/2011	2.5	0.4	4.46	14.00	17.17	43.86	29.15	6.73	7.40
NA03/Jul/2011	2.0	0.0	7.46	18.00	0.00	73.21	45.58	16.09	11.26
NA04/Jul/2011	1.7	0.0	9.62	25.00	0.00	82.30	49.90	19.75	12.47
NA05/Jul/2011	2.0	0.0	10.33	28.00	0.00	80.60	46.47	19.36	14.52
RV01/Jul/2011	2.7	0.0	6.34	12.00	0.00	57.39	42.59	12.62	2.05
RV02/Jul/2011	2.7	0.0	7.11	14.00	0.00	62.07	47.82	12.66	1.41
RV03/Jul/2011	1.7	0.0	7.72	12.00	0.00	78.00	60.88	10.36	6.48
RV04/Jul/2011	2.2	0.0	5.55	13.00	0.00	60.41	50.45	7.21	2.52
RV05/Jul/2011	2.2	0.0	5.45	18.00	0.00	59.61	45.87	7.34	6.24

NA: Nova América; RV: Rio Verde.

A total of 62 compounds were identified, however just the components that appear in most amounts or with more frequency were chosen for statistical analysis. The following compounds were selected: (*E*)-caryophyllene, α-humulene, α-zingibirene, hydroxytoluene butylated, caryophyllene alcohol, caryolan-8-ol, caryophyllene oxide, thujopsan-2-α-ol and *n*-heneicosane.

From the *stepwise* Multiple Regression, were obtained the following equations with the significant (p-value less than 0.05) variables (l=leaf and s=soil):

(*E*)-caryophyllene (%)=25.037-1.6615Nas-1.9111Nl+0.1424Mnl+33.514Col
(R^2=0.8113; R=0.9007) Equation 1

α-humulene (%)=8.3329-1.1731Nl+0.0084Fel+0.1271Mnl+23.217Col
(R^2=0.7856; R=0.8863) Equation 2

α-zingibirene (%)=-9.6726-12.833Als-0.4847Ss+10.96Sl+0.1053Mnl
(R^2=0.8725; R=0.9340) Equation 3

caryophyllenyl alcohol (%)= -2.3693+4.0911Mgs+3.8846(H+Al)
(R^2=0.5076; R=0.7124) Equation 4

caryophyllene oxide (%)=-4.4317+3.6199Mgl
(R^2=0.4132; R=0.6428) Equation 5

thujopsan-2-α-ol(%)=7.9808+2.4473(H+Al)-0.0262Fel
(R^2=0.4930; 0.7021) Equation 6

n-heneicosane (%) =15.311-0.8691Ss
(R^2=0.2584; R=0.5083) Equation 7

butylated hydrixytoluene = Significant coefficients were not found for this model caryolan-8-ol= Significant coefficients were not found for this model

Multiple Regression Analysis suggest that there the main factors capable to influence the levels of the compounds analyzed were: Als, Mgs, H+Al, Nas, Ss, Nl, Mnf, Col and Fel.

Multiple coefficient of determination (R^2) means the total proportion of the total variation that is explained by the overall regression model (Bowerman et al., 2005), when the R^2 is higher, better the model fits the data. Multiple correlation coefficient (R) is the positive squared root of R^2, and is the method in regression that is employed to look how far the relationship between two variables (Levinm & Rubin, 1994).

Equations 1, 2 and 3 showed a high explicability as can be seen by the higher R^2 values, which the main environmental factors that could predict the concentration of these compounds were foliar nutrients. The Equation 1 presents a very strong correlation (R>0.90) between (*E*)-caryophyllene and the set of variables (Nas, Nl, Mnl, Col), mainly the foliar nutrients were the factors capable to influenced the levels of (*E*)-caryophyllene (Piaw, 2006). These results suggests a strong association between (*E*)-caryophyllene levels and foliar nutrients. The compound (*E*)-caryophyllene stands out, due its many known pharmacological properties, such as: anti-inflammatory (Tambe et al., 1996), anticarcinogenic (Zheng et al., 1992), cytotoxic (Kubo et al., 1996), spasmolytic and local anesthetic (Cabo et al., 1986). Besides, this component may suffer oxidation when air exposure, resulting (*E*)-caryophyllene oxide, which present moderate allergenic activity (Skold et al., 2006).

The observed positive relationship between Mnl and (*E*)-caryophyllene, α-humulene and α-zingiberene is in agreement with the requirement of sesquiterpenes synthases for a divalent metal ion as cofactor. The observed positive relationship between Mnl and (*E*)-caryophyllene, α-humulene and zingiberene is in agreement with the requirement of sesquiterpenes synthases for a divalent metal ion as cofactor. The nutrient Mg also showed positive influence over caryophyllenyl alcohol and caryophyllene oxide, because Mg also is a divalent cation (Picaud et al., 2005; Duarte et al., 2010; 2012). The formation of sesquiterpenes in ginger (*Zingiber officinale* Roscoe; Zingiberaceae) is favored with Mg^{2+} as cofactor, which corroborates the results found in this paper (Picaud et al., 2006).

Cluster analysis employing Ward's method was also applied to the study of similarity of samples on the basis of constituent distribution and the samples showed a highly chemical variability within the essential oil from leaves of *S. jambos*. The dendrogram represented in Figure 1 suggests that the main factor that seems to influence the composition is the collection time, due of the formation of two clusters, one of them with samples of January (Cluster

Table 4. Levels of macronutrients (N_l, P_l, K_l, Ca_l, Mg_l, S_l in g/kg) and micronutrients (Cul, Fe_l, Mn_l, Zn_l in mg/kg) in the leaves of *Syzygium jambos* from each collection site in January 2010 to April 2011.

Sample	N	P	K	Ca	Mg	S	Cu	Fe	Mn	Zn
NA01/Jan/2011	12.00	1.00	10.00	5.00	1.70	1.00	6.00	310.00	34.00	20.00
NA02/Jan/2011	12.20	0.90	8.00	5.20	1.60	1.30	5.00	286.00	25.00	23.00
NA03/Jan/2011	12.00	1.20	10.60	4.90	1.50	1.00	5.00	276.00	24.00	22.00
NA04/Jan/2011	12.80	1.30	9.80	5.40	1.70	1.20	4.00	240.00	20.00	21.00
NA05/Jan/2011	12.00	1.20	8.40	4.70	2.20	1.10	7.00	243.00	18.00	19.00
RV01/Jan/2011	13.60	1.00	8.00	5.20	1.60	1.00	8.00	370.00	25.00	20.00
RV02/Jan/2011	12.50	1.00	8.00	5.50	1.60	1.10	5.00	305.00	22.00	21.00
RV03/Jan/2011	13.20	1.10	9.20	6.00	1.40	1.10	6.00	228.00	47.00	16.00
RV04/Jan/2011	14.00	1.40	9.60	6.20	1.60	1.20	6.00	202.00	61.00	16.00
RV05/Jan/2011	12.60	1.00	8.80	5.80	1.80	1.10	5.00	294.00	29.00	18.00
NA01/Jul/2011	14.00	1.00	7.60	5.20	1.70	1.20	4.00	241.00	29.00	11.00
NA02/Jul/2011	13.00	1.10	6.80	4.20	1.80	1.40	2.00	154.00	20.00	10.00
NA03/Jul/2011	12.60	1.00	8.00	4.40	1.60	1.60	3.00	367.00	19.00	11.00
NA04/Jul/2011	13.40	1.10	8.80	6.20	1.50	1.40	3.00	141.00	22.00	14.00
NA05/Jul/2011	12.80	1.20	8.60	6.50	1.60	1.50	3.00	148.00	24.00	14.00
RV01/Jul/2011	14.20	1.00	4.80	5.00	2.50	1.60	4.00	270.00	28.00	13.00
RV02/Jul/2011	14.40	1.10	6.40	6.00	1.80	1.80	4.00	363.00	22.00	14.00
RV03/Jul/2011	13.00	1.20	8.60	5.20	1.70	1.90	4.00	367.00	45.00	11.00
RV04/Jul/2011	13.20	1.20	8.00	5.40	1.60	1.60	6.00	345.00	58.00	11.00
RV05/Jul/2011	15.00	1.40	8.60	5.10	1.80	1.80	2.00	311.00	53.00	13.00

NA: Nova América; RV: Rio Verde

Table 5. Percentage of chemical constituents analyzed of samples of leaves from *Syzygium jambos*.

Sample	(*E*)-caryophyllene	α-humulene	α-zingibirene	butylated hydrixytoluene	caryophyllenyl alcohol	caryolan-8-ol	caryophyllene oxide	thujopsan-2-α-ol	*n*-heneicosane
NA01/Jan/2011	0	1.96	3.61	32.82	7.41	4.05	0.86	5.48	4.35
NA02/Jan/2011	0	2.07	4.4	12.75	11.36	7.67	0	5.76	5.51
NA03/Jan/2011	0	0.36	0.97	8.79	1.48	0.62	0	1.01	1.73
NA04/Jan/2011	0	0	3.69	6.08	9.89	4.55	1.13	5.87	14.15
NA05/Jan/2011	0	0	4.71	3.44	17.14	10.75	5.05	7.53	6.13
RV01/Jan/2011	0	1.02	2.69	10.84	16.54	9.62	2.7	9.42	15.35
RV02/Jan/2011	0	2.16	3.09	6.97	12.62	0	0	1.69	22.56
RV03/Jan/2011	0	3.51	5.02	10.45	11.44	6	1.64	6.24	15.13
RV04/Jan/2011	0	3.34	3.66	6.63	10.95	6.29	1.4	7.24	12.9
RV05/Jan/2011	0	2.82	3.54	8.36	11.68	6.49	1.67	8.29	11.35
NA01/Jul/2011	0	0.67	5.74	11.15	5.28	6.58	2.97	7.61	9.31
NA02/Jul/2011	0	0	1.25	5.79	9.31	8.09	3.26	9.45	17.75
NA03/Jul/2011	2.55	5.73	8.6	3.8	10.15	4.39	0	1.21	18
NA04/Jul/2011	4.49	2.36	5.06	15.61	15.3	9.05	1.88	12.19	13.33
NA05/Jul/2011	0	3.24	7.16	14.08	13.89	7.98	1.77	9.49	13.41
RV01/Jul/2011	2.76	0	3.92	10.99	11.52	7.55	3.51	9.96	4.43
RV02/Jul/2011	0	0.23	8.39	7.08	13.24	7.21	3.78	7.4	7.91
RV03/Jul/2011	3.26	5.57	12.27	4.29	9.1	4.95	1.22	5.49	12.97
RV04/Jul/2011	10.86	7.07	13.5	6.57	8.73	4.67	1.33	4.89	12.05
RV05/Jul/2011	9.46	6.49	17.73	6.75	7.59	4.04	1.36	4.71	15.25

I) and other group with samples of July (Cluster II). The Cerrado is characterized by two seasons: dry (April to September) and wet (October to March), which may have influenced the similarity profile of the samples in leaf essential oils from *S. jambos*, found in Cluster analysis (Santos et al., 2006).

The chemical variability in leaf essential oils from *S. jambos* determined by statistical analysis may reflect environmental influence on oil composition, although it may also have been promoted by genetic factors in cultivated samples (Duarte et al., 2010). This work suggests that there is an influence of environmental factors on the chemical composition of leaf essential oils of *S. jambos*, mainly foliar nutrients (N, Mn, Co, Fe, S and Mg) and soil nutrients (Na, Al, S and H+Al). The climate data seems had exerted a low influence due no significant correlation found in Multiple Regression Analysis, which is in agreement with Figure 1, where the dendrogram represents the localities as the main factor in the differences in samples.

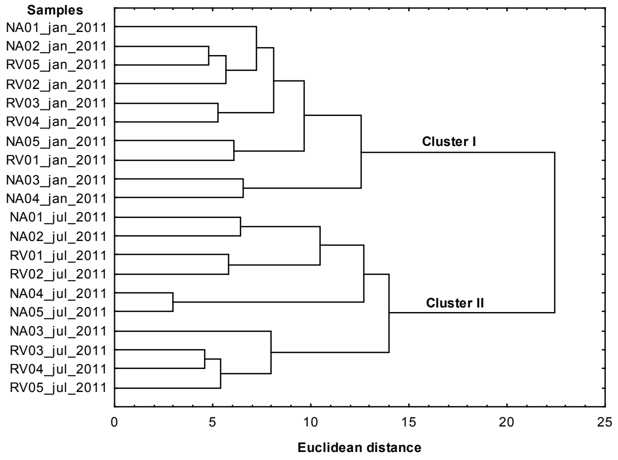

Figure 1. Dendrogram representing chemical composition similarity relationships among leaves of *S. jambos*, linking the climatic data, soil nutrients, foliar nutrients and essential oil composition according to Ward's variance minimization method.

Acknowledgment

We would like to thank the Brazilian government funding agencies CAPES and CNPq for the financial support.

Author's contributions

WPR contributed on interpretation of chromatograms and identification of structures; LLB contributed on statistical analysis, interpretation of results and drafting of the article; NMA in the extraction of essential oils; PHF contributed to essential oils analysis by GC-MS and JRP designed the study and drafting the manuscript.

References

Adams RP 2007. *Identification of Essential Oils Components by Gas Chromatography/Quadrupole Mass Spectroscopy.* Illiois, USA: Allured Publishing Corporation.

Amaral FMM, Ribeiro MNS, Barbosa-Filho JM, Reis AS, Nascimento FRF, Macedo RO 2006. Plants and chemical constituents with giardicidal activity. *Rev Bras Farmacogn 16*: 696-720.

Bergo CL, Mendonça HA, Silva MR 2005. Efeito da época e freqüência de corte de pimenta longa (*Piper hispidinervum* C. DC.) no rendimento de óleo essencial. *Acta Amazonica 35*: 111-117.

Bhargava UC, Westfall AB, Siehr DJ 1968. Preliminary pharmacology of ellagic acid from *Juglans nigra* (black walnut). *J Pharm Sci 57*: 1728-1732.

Bowerman BL, O'Connell RT, Koehler AB 2005. *Forecasting, time series and regression.* United States of America: Brooks/Cole Thomson Learning Inc.

Cabo J, Crespo ME, Jiménez J, Zarzuelo A 1986. The spasmolytic activity of various aromatic plants from the province of Granada. The activity of the major components of their essential oils. *Planta Med 20*: 213-218.

Cunha AP 2005. *Farmacognosia e Fitoquímica*. Lisboa: Fundação Calouste Gulbenkian.

Cruz GL 1979. *Dicionário das Plantas Ùteis do Brasil.* Rio de Janeiro: Civilização Brasileira S.A.

Fiuza TS, Rezende MH, Sabóia-Morais SMT, Bara MTF, Tresvenzol LMF, Paula JR 2008. Caracterização farmacognóstica das folhas de *Eugenia uniflora* L. (Myrtaceae). *Rev Eletronica Farm 5*: 21-31.

De Lima TCM, Klüeger PA, Pereira PA, Macedo-Neto WP, Morato GS, Farias MR 1998. Behavioral effects of crude and semi-purified extracts of *Syzygium cuminii* linn. skeels. *Phytother Res 12*: 488-493.

Di Stasi LC, Oliveira GP, Carvalhaes MA, Queiroz-Junior M, Tien OS, Kakinami SH, Reis MS 2002. Medicinal plants popularly used in the Brazilian Tropical Atlantic Forest. *Fitoterapia 73*: 69-91.

Djipa CD, Delmée M, Quetin-Leclercq J 2000. Antimicrobial activity of bark extracts of *Syzygium jambos* (L.) Alston (Myrtaceae). *J Ethnopharmacol 71*: 307-313.

Draper NR, Smith H 1981. *Applied regression analysis.* (2 ed.). New York: John Wiley.

Duarte AR, Santos SC, Seraphin JC, Ferri PH 2010. Environmental influence on phenols and essential oils of *Myrciaria cauliflora* leaves. *J Brazil Chem Soc 21*: 1672-1680.

Duarte AR, Santos SC, Seraphin JC, Ferri PH 2012. Influence of spatial, edaphic and genetic factors on phenols and essential oils of *Myrciaria cauliflora* fruits. *J Brazil Chem Soc 23*: 737-746.

Gondim ANS, Oliveira VR, Silva LR, Silva BA, Conde-Garcia EA 2006. Complete atrioventricular block on isolated guinea pig heart induced by an aqueous fraction obtained from *Psidium guajava* L. leaf. *Rev Bras Farmacogn 16*: 312-316.

Holetz FB, Pessini GL, Sanches NR, Cortez DAG, Nakamura CV, Filho BPD 2002. Screening of some plants used in Brazilian folk medicine for the treatment of infeccious diseases. *Mem I Oswaldo Cruz 97*: 1027-1031.

Inc SI 2002. Statistical Analysis System, Cary, NC, USA.

Kan WS 1987. *Manual of Medicinal Plants in Taiwan*. Taipei, Taiwan: Research Institute of Chinese Medicine.

Kelkar SM, Kaklij GS 1996. A simple two-step purification of antidiabetic compounds from *Eugenia jambolana* fruit-pulp: proteolytic resistance and other properties. *Phytomedicine 3*: 353-359.

Kim HM, Lee EH, Hong SH, Song HJ, Shin MK, Kim SH, Shin TY 1998. Effect of *Syzygium aromaticum* extract on immediate hypersensitivity in rats. *J Ethnopharmacol 60*: 125-131.

Kubo I, Chaudhuri SK, Kubo Y, Sanches Y, Ogura T, Saito T, Ishikawa H, Haraguchi H 1996. Cytotoxic and oxidative sesquiterpenoids from *Heterotheca inuloides. Planta Med 62*: 427-430.

Kurokawa M, Hozumi T, Basnet P, Nakano M, Kadota S, Namba T, Kawana T, Shiraki K 1998. Purification and characterization of Eugeniin as an antiherpesvirus compound from *Geum japonicum* and *Syzygium aromaticum. J Pharmacol Exp Therapeut 284*: 728-735.

Levinm RI, Rubin DS 1994. *Statistics for management*. (6 ed.) Englewood Cliffs, N.J.: Prentice Hall.

Lima MEL, Cordeiro I, Young MCM, Sobra MEG, Moreno PRH 2006. Antimicrobial activity of the essential oil from two specimens of *Pimenta pseudocaryophyllus* (Gomes) L.R. Landrum (Myrtaceae) native from São Paulo State - Brazil. *Pharmacologyonline 3*: 589-593.

Lorenzi H, Matos FJA 2002. *Plantas Medicinais no Brasil: Nativas e Exóticas.* Nova Odessa, Brazil: Instituto Plantarum.

Machado RB, Neto NBR, Pereira PG, Caldas E, Gonçalves DA, Santos NS, Tabor K, Steininger M 2004. *Estimativas de perda da área do cerrado brasileiro*. Brasília: Conservação Internacional.

MMA 2009. O Bioma Cerrado. Ministério do Meio Ambiente.

Moreira F 1978. *As Plantas Curam.* São Paulo: Hermus Liv. Ed. Ltda.

Muruganandan S, Srivastava K, Chandra S, Tandan SK, Lal J, Raviprakash V 2001. Anti-inflammatory activity of *Syzygium cumini* bark. *Fitoterapia 72*: 369-375.

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

Paula JAM, Ferri PH, Bara MTF, Tresvenzol LMF, Sá FAS, Paula JR 2011. Infraspecific chemical variability in the essential oils of *Pimenta pseudocaryophyllus* (Gomes) L.R. Landrum (Myrtaceae). *Biochem Syst Ecol 39*: 643-650.

Pessini GL, Holetz FB, Sanches NR, Cortez DAG, Dias Filho

BP, Nakamura CV 2003. Avaliação da atividade antibacteriana e antifúngica de extratos de plantas utilizados na medicina popular. *Rev Bras Farmacogn 13*: 21-24.

Piaw CY 2006. *Asas statistik penyelidikan* Kuala Lumpur: McGraw-Hill.

Picaud S, Olofsson ME, Brodelius M, Brodelius PE 2005. Expression, purification, and characterization of recombinant amorpha-4,11-diene synthase from *Artemisia annua* L. Arch. *Arch Biochem Biophys 436*: 215-226.

Picaud S, Olsson ME, Brodelius M, Brodelius PE 2006. Cloning, expression, purification and characterization of recombinant (+)-germacrene D synthase from *Zingiber officinale. Arch Biochem Biophys 452*: 17-28.

Potzernheim MCL, Bizzo HR, Vieira RF 2006. Análise dos óleos essenciais de três espécies de *Piper* coletadas na região do Distrito Federal (Cerrado) e comparação com óleos de plantas procedentes da região de Paraty, RJ (Mata Atlântica). *Rev Bras Farmacogn 16*: 246-251.

Rodrigues VEG, Carvalho DA 2001. *Levantamento etnobotânico de plantas medicinais no domínio do cerrado na região do alto Rio Grande - Minas Gerais.* Cienc Agrotec 25: 102-123.

Sá FAS, Borges LL, Paula JAM, Sampaio BL, Ferri PH, Paula JR 2012. Essential oils in aerial parts of *Myrcia tomentosa:* composition and variability. *Rev Bras Farmacogn 22*: 1233-1240.

Santos SC, Costa WF, Batista F, Santos LR, Ferri PH, Ferreira HD, Seraphin JC 2006. Seasonal variation tannins in barks of barbatimao. *Rev Bras Farmacogn 16*: 552-556.

Silva SC 2009. *Manual de análises químicas de solos, plantas e fertilizantes*. (2 ed.) Brasília: Embrapa Informação Tecnológica.

Simões CMO, Spitzer V 2004. Óleos voláteis. In: *Farmacognosia: da planta ao medicamento* (ed UFRGS/UFSC), Porto Alegre/Florianópolis, pp. 467-495.

Skold M, karlberg A, Matura A, Börje A 2006. The fragrance chemical β-caryophyllene - air oxidation and skin sensitization. *Food Chem Toxicol 44*: 538-545.

Slowing K, Carretero E, Villar A 1994a. Anti-inflammatory activity of leaf extract of *Eugenia jambos* in rats. *J Ethnopharmacol 43*: 9-11.

Slowing K, Carretero E, Villar A 1996. Anti-inflammatory compounds of *Eugenia jambos*. *Phytother Res 10*: 8126–8127.

Slowing K, Söllhuber M, Carretero E, Villar A 1994b. Flavonoid glycosides from *Eugenia jambos*. *Phytochemistry 37*: 255–258.

Souza LKH, Oliveira CMA, Ferri PH, Santos SC, Júnior JGO, Miranda ATB, Lião LM, Silva MRR 2002. Antifungal properties of Brazilian Cerrado plants. *Braz J Microbiol 33*: 247-249.

Stanely Mainzen P, Menon VP, Pari L 1998. Hypoglycaemic activity of *Syzigium cumini* seeds: effect on lipid peroxidation in alloxan diabetic rats. *J Ethnopharmacol 61*: 1-7.

Stat Soft I 2004. Statistica (data analysis software system).

Tambe Y, Tsujiuchi H, Honda G, Ikeshiro Y, Tanaka S 1996. Gastric cytoprotection of the non-steroidal antiinflamatory sesquiterpene, β-caryophillene. *Planta Med 62*: 469-470.

Van Den Dool H, Kratz PD 1963. Generalization of the retention index system including linear temperature programmed gas-liquid partition chromatography. *J Chromatog 11*: 463-471.

Ward JH 1963. Hierarchical grouping to optimize an objective function. *J Am Stat Assoc 58*: 66-103.

***Correspondence**

Leonardo Luiz Borges
Laboratório de Pesquisa de Produtos Naturais, Faculdade de Farmácia, Universidade Federal de Goiás
jrealino@farmacia.ufg.br
Tel. +55 62 3209 6183
Fax: +55 62 3209 6182