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CIRCADIAN AND SEASONAL VARIATION IN THE ESSENTIAL OIL FROM *VIROLA SURINAMENSIS* LEAVES

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Key Word Index—*Virola surinamensis*; Myristicaceae; essential oil; monoterpenes; sesquiterpenes; elemicin; circadian variation; seasonal variation.

Abstract—The essential oil from leaves of *Virola surinamensis* shows circadian variation in elemicin and in monoterpenes during the rainy season (February). The monoterpenes represents 50% of total volatile compounds during the dry season (June). Sesquiterpenes are predominant (50%) in the early rainy season (October). © 1997 Published by Elsevier Science Ltd

INTRODUCTION

Virola surinamensis is a myristicaceous tree that grows in the inundation plain of Amazonia (0° to 5° South) and produces seeds during the rainy season [1, 2]. Two seasons are well defined in this Amazonian region: the rainy season popularly known as 'inverno' (winter, October to March) and the dry season called 'verão' (summer, April to September). Seeds are viable shortly after ripening and are adapted to be dispersed by water or by toucans and macaws ('araras') [3]. Investigation of the chemical composition in different tissues of *Virola* species and its variation have revealed that flavones are replaced by lignans in fruits during the ripening process [4]. Our previous phytochemical investigation carried out on apolar extracts from seeds and leaves of *V. surinamensis* detected the occurrence of lignoids, propiophenone and γ-lactones [5, 6]. However, the essential oil of Brazilian Myristicaceae has never been investigated. This paper reports the composition of essential oil from leaves of *Virola surinamensis* collected at Museu Paraense Emílio Goeldi and describes its circadian variation during specific months of different seasons in a year.

RESULTS AND DISCUSSION

Evaluation of circadian and seasonal variations in the composition of the volatile constituents from *V. surinamensis* leaves was carried out on four samplings obtained from leaves collected at 6 a.m., 12 a.m., 6 p.m. and 9 p.m. during February, June and October. The essential oils were obtained by steam distillation and were analysed by GC and GC-mass spectroscopy. The identification of the components was based on their R_t and comparison with some authentic samples and also based on calculated Kovats index and its index combined with Adams retention time [7] (Table 1)

The effects of environmental factors on the accumulation of essential oils from *Mentha piperita*, *Ocimum basilicum*, *Coriandrum sativum*, *Anethum graveolens*, *Artemisia dracunculus* and *Foeniculum vulgare* were observed to be typical and specific to each species [8]. In our work the yield of essential oils from leaves was approximately constant (0.5%) in different seasons or during the day, but the relative composition of compound classes or individual components varied quite sharply as can be seen in Fig. 1. During February and June the relative level of sesquiterpenes was constant between 5–15%, but increased up to 50% during early phase of fruit development (October) (Fig. 1, Table 4). The major sesquiterpene component was identified as caryophyllene. High levels of caryophyllene on *Hymenae* sp. resin were correlated with effects on mortality or low insect herbivory [9]. The contents of monoterpenes and of elemicin were higher than the content of sesquiterpenes during February and June. Circadian variation was observed for contents of monoterpenes and of elemicin on a day in

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Table 1. Identification of essential oil components from *V. surinamensis* leaves by Kovats index (K_i) and retention time (R_t)

Compounds	K_i	K_i^*	R_t	R_t^*
α-Pinene	937	937	319	319
Camphene	950	951		
Sabinene	974	974		
β-Pinene	977	979		
Myrcene	990	991	406	408
α-Phellandrene	1006	1006		
α-Terpinene	1016	1017		
p-Cymene	1023	1024		
Limonene	1030	1029	480	481
cis-Ocimene	1037	1038		
trans-Ocimene	1047	1048		
γ-Terpinene	1058	1060	605	607
Terpinolene	1087	1088		
Linalool	1098	1099	634	631
4-Terpineol	1174	1175	856	855
p-Cymen-8-ol	1184	1182	484	481
α-Terpineol	1189	1188		
δ-Elemene	1338	1337		
β-Elemene	1393	1391		
Methyleugenol	1409	1402	1405	1407
Caryophyllene	1422	1418	1449	1450
α-Guaiene	1440	1437		
Germacrene D	1480	1479		
β-Selinene	1485	1484		
Valencene	1498	1491		
Elemol	1550	1547		
Elemicin	1560	1552		
Spathulenol	1578	1574		
Globulol	1583	1581		
γ-Eudesmol	1628	1628		
Torreyol	1644	1643		
α-Cadinol	1650	1651		

K_i-calculated.
K_i^*-calculated using Adams data.
R_t-observed.
R_t^*-standards.

February (Fig. 1, Table 2). The relative level of monoterpenes (28%) at 6 a.m. dropped to approximately half at noon (15%) and increased back to the same total content at 9 p.m. Exactly the opposite behaviour was observed in the elemicin content which reached a maximum of 26% at noon. In June, the content of monoterpenes increased to approximately 50%, which should suggest the importance of such volatile compounds during the onset of flowering. The limonene and α-pinene are the major constituents in the essential oil during this season (Fig. 1, Table 3).

EXPERIMENTAL

Plant Material. Leaves of *Virola surinamensis* were collected on 15th day of February, June and October of 1996 at 6 a.m., 12 a.m., 6 p.m. and 9 p.m. at Campus of Museu Paraense Emílio Goeldi (01° 27.133′ S; 048° 26.635′ W), Belém, Pará State, Brazil. A dry voucher sample (Lopes-034) has been deposited in the S.P.F-Herbário do Instituto de Biociências, Universidade de São Paulo.

Essential oils were obtained from dry leaves (200 g) by steam distillation and yielded 0.5 ± 0.05 ml. These essential oils were analysed by GC-MS (60–240° at 3° min. rate) on a GC Varian coupled to MS Finnigan INCOS-XL instrument using fused-silica capilar column ($30\ m \times 0.25$ mm i.d.) coated with DB-5. MS spectra were obtained using electron impact at 70 eV. Individual components were identified by comparison of both mass spectrum and their GC R_t data with those of authentic compounds previously analysed and stored in the library system. Other identifications were made by comparison of mass spectra and GC R_t with some standards and finally the comparison of calculated Kovats index using the R_t of the compounds and the R_t published by Adams [6].

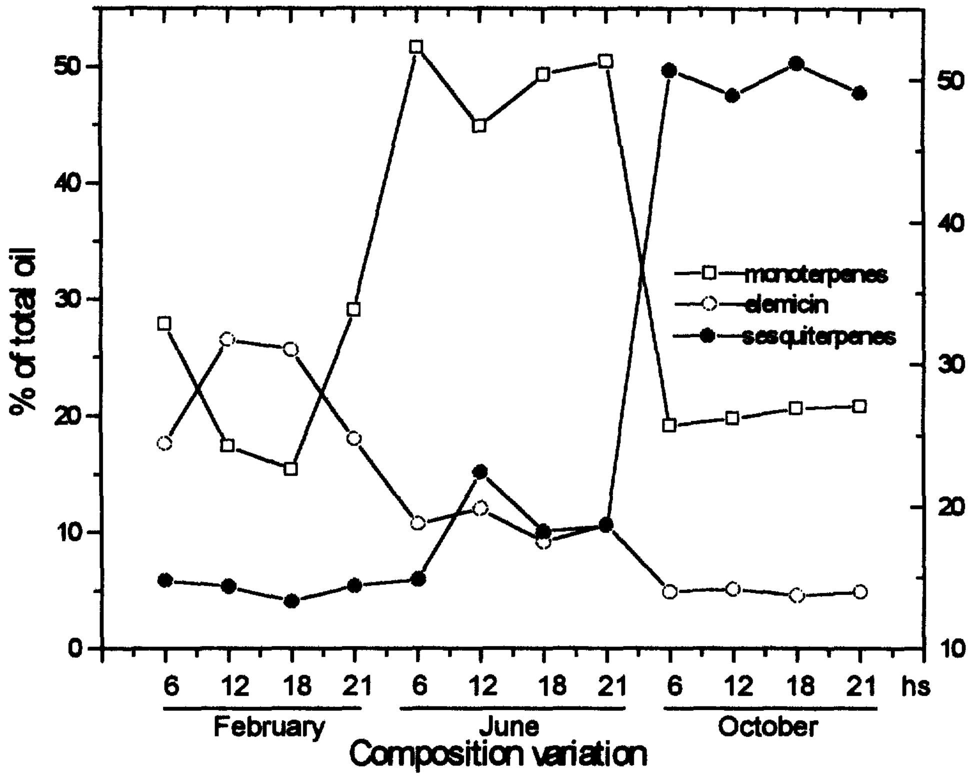

Fig. 1.

Table 2. Circadian variation on composition of essential oil from *V. surinamensis* leaves (February, 1996; % relative peak area)

Compounds	6 a.m.	12 a.m.	6 p.m.	9 p.m.
Monoterpenes				
α-Pinene	0.81			0.91
β-Pinene	1.10			1.06
Myrcene	1.78	0.73		1.95
p-Cymene	5.97	3.96	3.52	6.37
Limonene	19.29	12.59	11.87	19.85
Linalool	3.94	3.95	3.82	4.10
4-Terpineol		0.56	1.58	
p-Cymen-8-ol		0.50		
α-Terpineol	1.58	1.77		1.58
Sesquiterpenes				
δ-Elemene	0.67	0.71		0.69
β-Elemene	2.53	2.56	1.43	2.50
Caryophyllene	1.65	0.68	0.74	1.33
Germacrene D	1.52	1.46	1.70	1.55
β-Selinene	1.20	1.23	1.43	1.18
Valencene	7.12	7.34	7.10	7.29
Spathulenol	1.12	0.56		1.16
α-Cadinol	3.52	3.80	4.07	3.33
Phenylpropanoids				
Methyl eugenol		0.43		
Elemicin	17.65	26.51	25.66	18.03

Table 3. Circadian variation on composition of essential oil from *V. surinamensis* leaves (June, 1996; % relative peak area)

Compounds	6 a.m.	12 a.m.	6 p.m.	9 p.m.
Monoterpenes				
α-Pinene	13.26	12.25	14.44	14.19
β-Pinene	5.01	4.10	4.90	4.73
Myrcene	5.93	5.28	5.94	5.61
α-Phellandrene		0.43		
p-Cymene	5.80	4.55	5.22	5.20
Limonene	26.67	22.92	23.72	25.49
Linalool	1.22	1.40	1.47	1.22
4-Terpineol		0.24		
α-Terpineol	0.48	0.74	0.72	0.58
Sesquiterpenes				
δ-Elemene	0.40	0.36	0.40	0.37
β-Elemene	1.47	1.12	1.39	1.45
Caryophyllene	5.65	9.50	7.76	6.45
Germacrene D	0.86	0.88	0.91	1.02
β-Selinene	0.44	0.46	0.45	0.47
Valencene	5.83	10.15	8.30	7.65
α-Cadinol	1.96	1.62	0.76	3.10
Phenylpropanoids				
Methyl eugenol		0.28		
Elemicin	10.75	12.02	9.19	10.66

Table 4. Circadian variation on composition of essential oil from *V. surinamensis* leaves (October, 1996, % relative peak area)

Compounds	6 a.m.	12 a.m.	6 p.m.	9 p.m.
Monoterpenes				
α-Pinene	5.36	6.19	6.64	6.59
Camphene		0.12	0.16	0.17
Sabinene	0.11			
β-Pinene	2.65	2.92	3.12	3.01
Myrcene	3.17	3.28	3.41	3.35
α-Phellandrene	4.97	5.24	5.13	5.17
α-Terpinene	0.14	0.13	0.14	0.14
p-Cimene	0.23	0.24	0.26	0.26
Limonene	10.42	10.11	10.37	10.64
cis-Ocimene	0.15		0.08	
trans-Ocimene	0.19	0.09	0.11	0.11
γ-Terpinene	0.13		0.13	0.12
Terpinolene	0.36	0.29	0.30	0.31
Linalool	0.81	0.72	0.75	0.79
4-Terpineol	0.17	0.11	0.14	0.14
α-Terpineol		0.36	0.47	0.44
Sesquiterpenes				
δ-Elemene	1.64	1.51	1.56	1.52
β-Elemene	1.71	1.81	1.84	1.89
Caryophyllene	23.95	20.40	24.50	22.26
α-Guaiene	0.08		0.09	0.08
Valencene	24.24	25.44	24.11	23.95
Elemol		0.06		
Globulol		0.21		
γ-Eudesmol	0.18	0.25	0.21	0.24
Torreyol	0.10	0.16		
α-Cadinol	0.78	1.28	0.71	1.02
Phenylpropanoids				
Methyleugenol	0.20			0.12
Elemicin	4.91	5.15	4.60	4.95

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