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Chemical characterization of the essential oil from patchouli accessions harvested over four seasons

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ABSTRACT

The genus *Pogostemon* of the Lamiaceae family includes several species known for their medicinal and aromatic properties. The species *P. cablin* is especially notable because the essential oil extracted from its leaves is internationally important and valuable, principally for the perfume and cosmetic industries. Because multiple factors can affect the chemical composition of the essential oil, the aim of this work was to evaluate the chemical variations in the essential oils of nine *Pogostemon* accessions harvested over four seasons. Two *Pogostemon* accessions (*P. heyneanus*, but received as *P. cablin*) and seven *P. cablin* accessions from different sources were evaluated. The transplants were planted in January 2008, and the harvests were conducted in May, August, and November 2008 and February 2009. The chemical composition of the essential oils was evaluated quantitatively and qualitatively. Patchoulol was the major compound from the four harvests of all of the *P. cablin* accessions. The principal compound from the accessions POG-001 and POG-006 was β-pinene. Two clusters were detected by multivariate analyses of the four harvests. Cluster I was formed by the accessions POG-001 and POG-006 (*P. heyneanus*), and Cluster II was formed by the accessions of *P. cablin* (POG-002, POG-014, POG-015, POG-016, POG-019, POG-021, and POG-022).

1. Introduction

The Lamiaceae family is composed of herbaceous or shrubby, rarely woody, perennial or annual species with an aromatic and even woody fragrance, including diverse species of value in culinary and medicinal applications, as well as in the manufacture of perfumes and cosmetics (Guenther, 1972).

The species *Pogostemon cablin* (Blanco) Benth., a member of the Lamiaceae family, is notable for its various applications as a reducer of appetite, water retention, and inflammation; a cell rejuvenator and antiseptic; an aphrodisiac; an aid in the treatment of acne, eczema, nervousness, depression, and insomnia; a fungicide; an insecticide; an aid in combating menstrual problems; an antirheumatic; a treatment for headaches; and a tranquilizer, sedative, and hypotensive (Zhu et al., 2003; Pavela, 2005; Oyen and Dung, 2006; Khare, 2007; Wei and Shibamoto, 2007).

According to several studies, the essential oil of patchouli (*P. cablin*) most frequently contains the following compounds: β-elemene, varying from 0.24 to 1.3%, β-patchoulene (0.94–12.12%), β-caryophyllene (1.88–5.14%), α-patchoulene (2.3–20.9%), α-guaiene (3.17–22.2%), seychellene (4.73–8.94%), α-bulnesene (9.86–20.3%), and patchoulol (17.5–54.31%) (Singh et al., 2002; Zhu et al., 2003; Bure and Sellier, 2004; Milchard et al., 2004; Anonis, 2006; Hu et al., 2006; Lawrence, 2007; Tsai et al., 2007; Wei and Shibamoto, 2007).

As the main component of the essential oil of patchouli, patchoulol is important for the duration of the odor of the patchouli essential oil and is frequently used as an indicator of essential oil quality (Anonis, 2006).

Species of the Lamiaceae family exhibit great chemical variability in their essential oils and can exist as chemotypes, plants of the same species that are phenotypically identical but chemically distinct (Vieira and Costa, 2006). Chemical analysis of the oil of *P. cablin* has revealed the existence of two chemotypes, the pogostone type and the patchoulol type (Hu et al., 2006). The existence of these two chemotypes was also observed by Luo et al. (2003) and confirmed by molecular testing. The species *Pogostemon heyneanus* Benth. is also called patchouli, but its essential oil is of lower industrial interest, because of the lower patchoulol content (Oyen and Dung, 2006).

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Table 1
Accessions of patchouli (*P. heyneanus* and *P. cablin*) from the Active Germplasm Bank of the Federal University of Sergipe.

Accession code	Scientific name	Origin	Voucher number
POG-001	*Pogostemon heyneanus*	"Centro Pluridisciplinar de Pesquisas Químicas, Biológicas e Agrícolas – Universidade Federal de Paraná", Campinas – SP – Brazil	13169
POG-002	*Pogostemon cablin*	Free market, São Paulo – SP – Brazil	13170
POG-006	*Pogostemon heyneanus*	Richters company, Canada	13171
POG-014	*Pogostemon cablin*	Free market, Belo Horizonte – MG – Brazil	13172
POG-015	*Pogostemon cablin*	Free market, São Cristóvão – SE – Brazil	13173
POG-016	*Pogostemon cablin*	"Embrapa Recursos Genéticos e Biotecnologia", Brasília – DF – Brazil	13174
POG-019	*Pogostemon cablin*	"Universidade Federal de Paraná", Curitiba – PR – Brazil	13176
POG-021	*Pogostemon cablin*	Growmore Ltd. company, India	13177
POG-022	*Pogostemon cablin*	Free market, Belém – PA – Brazil	13178

The chemical characterization of accessions of plants that produce essential oils and are maintained in germplasm banks, is important for identifying higher quality genotypes, because of the better adaptation to the region of cultivation.

Because of the existence of chemotypes and other factors, such as drying of leaves, season and time of harvest (Blank et al., 2005, 2007; Carvalho Filho et al., 2006), that may influence the production of essential oils, the available accessions must be well-characterized. Therefore, the objective of this study was to evaluate the chemical variation in the essential oils of the accessions of patchouli from the Active Germplasm Bank of the Federal University of Sergipe, harvested over four seasons.

2. Materials and methods

2.1. Plant material and crop experimental design

The experiment was conducted at the Research Farm "Campus Rural da UFS", located in São Cristóvão-SE, latitude 11°00′S and longitude 37°12′W. Plants were evaluated from nine accessions of patchouli from the Active Germplasm Bank (AGB) cultivated in field at the Federal University of Sergipe (UFS) (Table 1). From these accessions, two were identified as *P. heyneanus* and seven were *P. cablin*.

By analyzing the material deposited in the Herbarium of the Federal University of Sergipe (ASE), we observed differences in the morphological characteristics of accessions POG-001 and POG-006 (*P. heyneanus*) when compared to the other accessions analyzed (*P. cablin*). Morphological differences found between the two species included differences in the stem, branches, leaves, and trichomes. The stems of individuals of *P. heyneanus* have a different coloration and fewer trichomes than the stems of *P. cablin*, which contained a large amount of trichomes in all individuals analyzed. The leaves of *P. heyneanus* have a different length and format than those of *P. cablin* and are often ovoid and 1–3 × 1.5–3 cm in size, and the trichomes are most evident in the veins of the adaxial face. *P. cablin* has a larger leaf blade (3–8 × 2–7 cm) and a higher intensity of trichomes on the blade on both sides, with no differentiation as described above for the other species. In addition to their morphological differences, *P. cablin* does not flourish in the region while *P. heyneanus* does.

Plants from the AGB were produced in expanded polystyrene trays with 72 cells from cuttings from stock plants grown at the Research Farm "Campus Rural da UFS". The substrate was composed of coconut coir and vermiculite in a 1:1 ratio, plus 1 g dm^{-3} limestone and 2 g dm^{-3} NPK (3-12-6). The trays were kept in a greenhouse covered by a 50% shade screen with intermittent nebulization until the plantlets were transplanted to the experimental area.

To prepare the soil of the experimental area, 200 g m^{-2} of limestone was used to raise the base saturation to 80%. The soil was fertilized by applying 6 kg m^{-2} of cattle manure and 200 g m^{-2} of NPK (3-12-6). Drip irrigation was used, and the planting area was covered by plastic mulch with two colors: black underside and white topside.

Table 2
Main volatile components (% calculated by GC-FID) in essential oil of the first harvest of accessions of *P. heyneanus* and *P. cablin* from the Active Germplasm Bank of the Federal University of Sergipe.

Compound	KI	POG-001	POG-002	POG-006	POG-014	POG-015	POG-016	POG-019	POG-021	POG-022
α-Pinene	939	17.23 a	0.00 b	21.40 a	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b
β-Pinene	979	33.25 a	0.00 b	40.13 a	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b
Limonene	1029	1.53 a	0.00 b	1.59 a	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b
Acetophenone	1065	17.28 a	0.00 b	12.90 a	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b
β-Patchoulene	1380	0.67 b	2.84 a	0.61 b	2.30 a	2.68 a	1.91 a	1.76 a	2.15 a	2.51 a
β-Elemene	1390	0.00 b	0.91 a	0.00 b	0.72 a	0.80 a	0.59 a	0.63 a	0.87 a	0.85 a
β-Caryophyllene	1419	3.37 a	3.24 a	3.28 a	2.41 b	2.77 a	2.07 b	1.95 b	2.95 a	3.04 a
α-Guaiene	1439	1.64 c	9.53 a	1.49 c	7.43 b	9.15 a	6.04 b	5.81 b	8.98 a	9.32 a
Seychellene	1446	1.17 b	6.50 a	1.05 b	5.01 a	5.92 a	4.07 a	4.01 a	5.54 a	6.08 a
α-Humulene	1454	0.46 b	0.60 a	0.42 b	0.63 a	0.70 a	0.39 b	0.49 b	0.56 a	0.54 a
α-Patchoulene	1456	0.79 b	3.91 a	0.75 b	3.14 a	2.72 a	2.33 a	2.16 a	3.44 a	2.96 a
α-Bulnesene	1509	0.97 b	13.00 a	0.94 b	10.32 a	11.48 a	8.72 a	8.93 a	12.14 a	11.90 a
(E)nerolidol	1563	9.62 a	0.00 c	6.65 b	0.00 c	0.00 c	0.00 c	0.00 c	0.00 c	0.00 c
Caryophyllene oxide	1583	0.55 a	0.00 b	0.38 a	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b	0.00 b
β-Atlantol	1608	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a
Pogostol	1653	0.44 b	3.25 a	0.33 b	4.18 a	3.81 a	4.63 a	4.52 a	3.57 a	3.59 a
Patchoulol	1658	9.04 b	47.26 a	6.28 b	55.25 a	55.69 a	60.52 a	62.97 a	50.50 a	55.51 a
EO content (%)		1.84 b	1.59 b	1.05 c	1.75 b	1.18 c	2.44 a	1.90 b	1.72 b	1.56 b
Dry weight of leaves (g $plant^{-1}$)		37.30 a	31.70 a	33.20 a	34.29 a	32.05 a	36.10 a	31.92 a	14.30 b	16.92 b

Means followed by the same letter in the rows, do not differ between themselves by the Skott–Knott test ($p \leq 0.05$).
KI = Kovats Index.