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Research Article

Essential oil composition of *Siparuna lepidota* (Kunth) A. DC. (Siparunaceae) from Ecuador

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Abstract

The essential oil from the leaves of *Siparuna lepidota* (Kunth) A. DC. was collected from cultivated species in Guayaquil, Ecuador. The oil was obtained by steam distillation and analyzed using GC/MS and GC/FID and it revealed a high content of monoterpene hydrocarbons (83.7%). The essential oil was rich in limonene (71.5%), with other prominent compounds such as β-pinene (5.2%), α-pinene (2.8%), myrcene (1.7%), α-terpinolene (2.3%), δ-cadinene (2.1%), α-copaene (1.7%), and (E)-cadina-1,4-diene (1.4%). To the best of the authors' knowledge, this study is the first to present the chemical composition of *Siparuna lepidota* essential oil, providing fundamental data for future research into its ethnobotanical uses and biological properties.

1. Introduction

The Siparunaceae family contains two genera: *Glossocalyx* and *Siparuna*. While *Glossocalyx* is comprised of only a few species in West Africa, *Siparuna* has about 58 species with a range that includes Central America and the West Indies and reaches throughout northern South America to Paraguay [1-3]. Most of these species are shrubs, with some trees, and are found at elevations from sea-level to 3800 m and in a wide variety of habitats, spanning lowland rain forest, montane forests, subpáramo scrub forest, lower elfin forest, and grassland gallery forest [1].

Some species of the *Siparuna* genus are noted for their characteristic, strong, citrus aroma given off by the leaves, fruit, and bark [1, 4]. *Siparuna spp.* are notable not only for their large and abundant oil-containing cells in each of their plant parts but also for their rich composition of biologically active compounds, including benzylisoquinoline alkaloids, sesquiterpenes, and flavonoids [1, 4]. *Siparuna* species have been traditionally utilized by native peoples throughout South and Central America for various medicinal purposes. They are used in traditional medicine as antimalarials and febrifuges, with practices including the boiling of macerated leaves to create medicinal baths aimed at alleviating fever, cold symptoms, and rheumatism [1, 5]. The research also highlighted the extracts from the leaves of *S. grandiflora* (referred to as *S. tonduziana*), *S. pauciflora*, and *S. thecaphora* demonstrate activities against the malaria parasite, which supports their use in traditional antimalarial therapies [1]. Additionally, applications of *Siparuna spp.* leaves or bark in poultices for the treatment of snake bites and minor wounds are documented, as is the use by the Ecuadorian Quichua of heated bark from species such as *S. sessiliflora* and *S. thecaphora* for hastening the recovery from herpes sores [1].

Ecuador, a country with some of the greatest biodiversity in the world [6], has approximately 40 species of the *Siparuna* genus [7], and *Siparuna lepidota* is one such species. *S. lepidota* is a shrub or treelet that can be 3-10 m in height, with yellow flowers, and small round fruit that is reddish-purple when ripe [1]. Common names include chiri guayusa, guayusa de montaña, and limoncello [1, 8]. While the ethnobotanical and medicinal uses of *Siparuna lepidota* are not extensively documented, it was reported that the fruits were decocted in water to produce an extract used for treating stomach colics, in medicinal baths, and to impart a lemon flavor to beverages [1]. Additionally, the juice extracted from the leaves of this species is applied topically as a remedy for ear pain [8].

A few species of *Siparuna*, such as *S. echinata*, *S. guianensis*, *S. muricata*, *S. thecaphora*, *S. cymosa*, and *S. brasiliensis* have been studied for their essential oil content [9-15]. Little research exists to elucidate the chemical properties of *Siparuna lepidota* essential oil, which may substantiate its traditional uses. To the authors' best knowledge, the essential oil of *S. lepidota* has not been previously reported. In the present study, GC/MS and GC/FID analytical techniques are used to establish the chemical profile of this essential oil from cultivated samples in Guayaquil, Ecuador, providing a foundation for future research.

2. Materials and methods

Siparuna lepidota leaves (Fig. 1) were collected in August 2023 from cultivated populations in Guayaquil, Ecuador (2°16'36.8"S 80°03'56.1"W). Plant material was stored in a shaded location for 2 days before distillation. A representative voucher sample of *S. lepidota* was deposited in the herbarium Universidad de Guayaquil (13.553GUAY). Steam distillation was carried out for 2 hours. The essential oil obtained was separated by a cooled condenser, collected, filtered,

Figure 1. Botanical illustration of *Siparuna lepidota* plant part used in the study, namely the leaves. Illustrated by Rick Simonson, Science Lab Studios, Inc. (Kearney, NE, USA).

and stored in sealed amber vials at room temperature (25 °C) until analysis. The essential oil yield was calculated as the ratio of the essential oil volume (mL) to the plant material mass (kg) before the distillation process.

Essential oil was analyzed, and volatile compounds were identified by GC/MS using an Agilent 7890BGC/5977B MSD (Agilent Technologies, Santa Clara, CA, USA) and Agilent J&W DB-5, 60 m × 0.25 mm,0.25 µm film thickness, fused silica capillary column. Operating conditions: 0.2 µL of the sample, 25:1 split ratio, initial oven temperature of 60 °C with an initial hold time of 2 min, oven ramp rate of 4.0 °C per minute to 245 °C with a hold time of 5 min, helium carrier gas. The electron ionization energy was 70 eV, scan range 35–550 amu, scan rate 2.4 scans per second, source temperature 230 °C, and quadrupole temperature 150 °C. Volatile compounds were identified using the Adams volatile oil library [16] using Chemstation library search in conjunction with retention indices. Volatile compounds were quantified and reported as a relative area percent by GC/FID using an Agilent 7890B and Agilent J&WDB-5, 60 m × 0.25 mm, 0.25 µm film thickness, fused silica capillary column. Operating conditions: 0.1 µL of sample, 25:1 split injection, initial oven temperature at 40 °C with an initial hold time of 2 min, oven ramp rate of 3.0 °C per minute to 250 °C with a hold time of 3 min, helium carrier gas. Essential oil samples were analyzed in triplicate by GC/FID to ensure repeatability (standard deviation < 1 for all compounds).

3. Results and discussion

The essential oil yield of *Siparuna lepidota* was 13.3 mL/kg, and the chemical profile is detailed in Table 1, revealing this essential oil is rich in monoterpene hydrocarbons (83.7%).

Table 1. Chemical profile of *S. lepidota* essential oil determined by GC/FID.

KI	Compound Name	Area percentage (%)
932	α-Pinene	2.8
969	Sabinene	0.2
974	β-Pinene	5.2
988	Myrcene	1.7
1024	Limonene	71.5
1086	α-Terpinolene	2.3
1137	(E)-Limonene oxide	0.3
1179	p-Cymen-8-ol	0.1
1186	α-Terpineol	0.1
1235	Neral	0.2
1249	Geraniol	0.2
1264	Geranial	0.3
1348	α-Cubebene	1.1
1374	α-Copaene	1.7
1387	β-Cubebene	1.3
1417	(E)-Caryophyllene	0.7
1448	cis-Muurola-3,5-diene	1.3
1452	α-Humulene	0.6
1458	Allo-aromadendrene	0.5
1478	γ-Muurolene	0.3
1480	Germacrene D	1.1
1493	(E)-Muurola-4(14),5-diene	0.9
1500	Bicyclogermacrene	0.5
1521	(E)-Calamenene	0.5
1522	δ-Cadinene	2.1
1533	(E)-Cadina-1,4-diene	1.4
	Compound Classes	
	Monoterpene hydrocarbons	83.7
	Oxygenated monoterpenes	1.2
	Sesquiterpene hydrocarbons	14.0
	Total identified compounds	**98.9**

Note: Essential oil sample was analyzed in triplicate to ensure repeatability (standard deviation < 1). Unidentified compounds of less than 0.5% are not included. KI is the Kovat's Index previously calculated by Robert Adams using a linear calculation on a DB-5 column [16]. Relative area percent was determined by GC/FID.

Twenty-seven compounds of *S. lepidota* essential oil were identified. Limonene was the most abundant component in the essential oil (71.5%). Other notable monoterpenes include α-pinene (2.8%), β-pinene (5.2%), myrcene (1.7%), and α-terpinolene (2.3%), The second most abundant group in the essential oil were the sesquiterpene hydrocarbons, including compounds such as δ-cadinene (2.1%), α-copaene (1.7%), and (E)-cadina-1,4-diene (1.4%).

Reviewing the chemical compositions of essential oils from species within the same genus, previous studies have demonstrated that the essential oil of *S. echinata* is characterized by a high content of monoterpene hydrocarbons, with α-pinene, β-pinene, β-myrcene, and limonene being the major compounds, the latter constituting 10.0% [9]. Similarly, the essential oil of *S. muricata* is dominated by hydrocarbon monoterpenes, with α-pinene as the principal component and a significant amount of limonene (8.7%) [7]. In contrast to the previously mentioned species, which is characterized by major monoterpenes, the essential oil of *S. guianensis* leaves has been reported to contain major compounds such as the sesquiterpenes γ-muurolene, curzerene, and curzerenone [10]. Furthermore, distinct chemotypes of *S. guianensis* have been identified including one with α-bisabolol as the major compound, and the other with germacrene D as the predominant component [11]. For *S. thecaphora*, the major compounds identified in its essential oil are spathulenol, and 2-tridecanone [12]. Additionally, the essential oil of *S. cymosa* is primarily composed of α-bisabolol [14], while *S. brasiliensis* is characterized by γ-muurolene and 2-undecanone as its major components [15].

Comparing the chemical composition of *Siparuna lepidota* essential oil with previously reported species within the genus *Siparuna*, both similarities and notable differences are observed. In our study, limonene was the most abundant component (71.5%), being significantly higher than the limonene content reported in *S. muricata* (8.7%) and in *S. echinate* (10.0%) [7, 9]. Additionally, while *S. lepidota* essential oil contains α-pinene, β-pinene, and myrcene, these monoterpenes are also present in *S. echinata* and *S. muricata*, though in different proportions, highlighting the unique profile of *S. lepidota* species essential oil, which is particularly rich in limonene. These differences in chemical composition not only underscore the chemical diversity within the genus *Siparuna*, but also could have significant implications for their uses in the fragrance and aromatherapy industries, where limonene-rich essential oils are valued for their citrus-like properties and potential therapeutic benefits.

The most abundant chemical constituents of essential oils generally dictate their bioactivities [17, 18]. Limonene, the most abundant compound present in the essential oil of *S. lepidota*, has been studied for its biological activities. Limonene has been shown to have antiproliferative, apoptotic, and anti-carcinogenic properties [19, 20]. Many studies have revealed that limonene provides antioxidant properties [21]. Also, this compound has been reported as an effective anti-inflammatory [21], which could be related to the traditional use of *S. lepidota* as a remedy for ear pain. Future studies need to be conducted to investigate the biological activities of *S. lepidota* essential oil.

4. Conclusions

To the best of the authors' knowledge, this study is the first to present the chemical composition of *Siparuna lepidota* essential oil. The essential oil of *S. lepidota*, with a high limonene concentration (71.5%), indicates significant potential for therapeutic and commercial applications. Although it shares some monoterpenes with other *Siparuna* species, the high limonene content is particularly notable. This suggests antioxidant and anti-inflammatory benefits, consistent with traditional use, and highlights potential applications in the pharmaceutical and cosmetic industries. Further studies are recommended to evaluate its biological activities and explore industrial and medicinal applications.

Authors' contributions

Conceptualization, C.P.; Methodology, C.P., A.A.; Software, C.P., A.A., T.O.; Validation, C.P.; Formal analysis (GC/MS, GC/FID), C.P., A.A., T.M.W., T.O.; Investigation, C.P., A.A.; Resources, C.P., N.C., E.C., O.P.; Data curation, C.P., A.A.; Writing– original draft, C.P. A.A.; Writing–review & editing, C.P., A.A., N.C., T.O., E.C., O.P., T.M.W.

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Availability of data and materials

All data will be made available on request according to the journal policy.

Conflicts of interest

The authors declare no conflict of interest. The funding entity had no role in the design of the study, in the collection, analysis, or interpretation of data, in the writing of the manuscript, or in the decision to publish the results.

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