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Vitex doniana L. Growing in Southwestern Nigeria: Leaf Essential Oil Composition and Antimicrobial Activity

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Abstract

Introduction: *Vitex doniana* is used in traditional medicine for treatment of several ailments, including conjunctivitis, skin rashes due to measles or chickenpox, respiratory infections, and abdominal disorders and diarrhea. **Methods:** The leaf essential oil of *V. doniana* from southwestern Nigeria was obtained by hydrodistillation and analyzed by gas chromatography-mass spectrometry. The essential oil was screened for antimicrobial activity against a panel of pathogenic bacteria and fungi. **Results:** Major components in the essential oil were (*E*)-β-caryophyllene (12.34%), phytone (9.73%), phytol (16.87%), and incensyl acetate (23.57%). **Conclusion:** The essential oil showed notable antifungal activity against *Aspergillus niger* (MIC 78.1 μg/mL) and should be considered as an alternative or complementary option for antifungal therapy.

Keywords

black plum, essential oil, β-caryophyllene, phytone, phytol, incensyl acetate, antibacterial, antifungal

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Introduction

The genus *Vitex* (Lamiaceae) is made up of 233 recognized species,[1] dominated by trees and shrubs, found in tropical and sub-tropical regions.[2,3] Common species used in ethnobotanical medicine include *V. agnus-castus* L.,[4] *V. negundo* L.,[5] *V. rotundifolia* L.,[6] and *V. trifolia* L.[7]

Vitex doniana Sweet (black plum) is a deciduous flowering tree growing up to 20 m height in the forests of coastal tropical west Africa. It is recognized with long-stalked leaves rounded at the apex with a glabrous leaflet and edible fruits.[8] *V. doniana* is known locally as "Oori nla" in Yoruba, "Dinya" in Hausa and "Ucha Koro" in Igbo. In folk medicine, *V. doniana* is used for treatment of several ailments, for example conjunctivitis, skin rashes due to measles or chickenpox, respiratory infections, and abdominal disorder and diarrhea.[9–11] Antimicrobial potency of *V. doniana* has been reported against several bacteria, including methicillin resistant *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Shigella dysenteriae*, *Pseudomonas aeruginosa*, and *Salmonella typhi* among others.[12–17] Hydroxycinnamic acid, saponins, allicins, flavonoids, and terpenoids have been identified in leaf extracts of *V. doniana*.[18] This present report is aimed at the characterization of the components of the essential oil of *V. doniana* and its antimicrobial activity for future exploitation in pharmaceutical applications.

Results and Discussion

Essential Oil Composition

Hydrodistillation of *V. doniana* leaves gave a pale-yellow essential oil in 0.65% (v/w) yield. The essential oil was analyzed by gas chromatography-mass spectrometry (GC-MS). A total of 21 compounds were identified in the essential oil (Table 1), which was predominantly (46.75%) oxygenated diterpenoids with phytol (16.87%) and incensyl acetate (23.57%) as the dominant components. Sesquiterpene hydrocarbons (20.31%) also made up a large percentage of the composition with (*E*)-caryophyllene (12.34%), α-humulene (3.71%), and *ar*-curcumene (3.22%).

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Notably, monoterpene hydrocarbons were not observed and linalool was the only oxygenated monoterpenoid in only 2.92%. Other constituents in high concentration were phytone (9.73%) and *p*-vinylanisole (5.82%).

A previous report on *V. doniana* essential oil also showed phytol and (*E*)-β-caryophyllene to be major components.[23] In contrast, however, β-phellandrene (31.3%) was the dominant component in the previous study, but this compound was not detected in the present study. Likewise, incensyl acetate (23.57%) and phytone (9.73%) were major components in the present study, but were not observed in the previous work. Variation in the composition of the present and the previous study may be attributed to climatic, geographical, and environmental factors.[24,25] Interestingly, *Vitex ajugifolia* and *Vitex pinnata* leaf essential oils also showed a paucity of monoterpenoids, while *Vitex trifolia* essential oils were dominated by monoterpenoids.[26] Furthermore, *Vitex negundo* essential oils from Vietnam have shown large variations in chemical composition depending on geographical location.[27]

Table 1. Chemical Composition of the Leaf Essential Oil of *Vitex doniana* From Southwestern Nigeria.

			Percent Composition[a]	
RI_{calc}	RI_{db}	Compound	Ave.	St. Dev.
1100	1101	Linalool	2.92	0.21
1152	1153	*p*-Vinylanisole	5.82	0.18
1377	1375	α-Copaene	1.05	0.29
1378	1379	(*E*)-β-Damascenone	0.48	0.13
1420	1417	(*E*)-β-Caryophyllene	12.34	0.41
1447	1447	Geranyl acetone	1.05	0.16
1456	1454	α-Humulene	3.71	0.12
1477	1481	(*E*)-β-Ionone	1.88	0.35
1481	1481	*ar*-Curcumene	3.22	0.43
1561	1560	(*E*)-Nerolidol	1.57	0.19
1582	1577	Caryophyllene oxide	3.21	0.25
1610	1613	Humulene epoxide II	1.56	0.15
1676	1679	Hexyl salicylate	1.29	0.19
1743	1746	2-Hexyl-(*E*)-cinnamaldehyde	1.03	0.46
1837	1836	Neophytadiene	1.17	0.56
1841	1841	Phytone	9.73	0.53
1867	1872	Benzyl salicylate	2.43	0.97
1946	1947	Isophytol	1.42	0.12
2108	2106	Phytol	16.87	3.81
2145	2149	Incensyl acetate	23.57	0.72
2148	2143	Serratol	3.70	0.32
		Monoterpene hydrocarbons	0.00	
		Oxygenated monoterpenoids	2.92	
		Sesquiterpene hydrocarbons	20.31	
		Oxygenated sesquiterpenoids	6.33	
		Diterpenoids	46.73	
		Benzenoid aromatics	10.56	
		Others	13.14	
		Total identified	100.00	

RI_{calc}, retention index calculated with respect to a homologous series of *n*-alkanes on a ZB-5 ms column; RI_{db}, reference retention index from the databases.[19–22]

[a]Three measurements.

Antibacterial and Antifungal Activity

The *V. doniana* essential oil and 3 essential oil components were assayed for antibacterial and antifungal activity using the microbroth dilution technique against a panel of pathogenic bacteria, namely *Bacillus cereus*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Streptococcus pyogenes*; and pathogenic fungi *Aspergillus fumigatus*, *Aspergillus niger*, *Cryptococcus neoformans*, *Microsporum canis*, *Microsporum gypseum*, *Trichophyton mentagrophytes*, *Trichophyton rubrum*, and *Candida albicans* (see Table 2).

The antimicrobial screening of *V. doniana* leaf essential oil revealed a broad range of activities and inhibited 14 microorganisms with minimum inhibitory concentration (MIC) ranging from 78.1 to 1250 μg/mL. Holetz and co-workers have defined antimicrobial activities of botanicals as good activity (MIC < 100 μg/mL), moderate activity (MIC 100-500 μg/mL), weak activity (MIC 500-1000 μg/mL), and inactive (MIC > 1000 μg/mL).[28] Thus, *V. doniana* essential oil was strongly active against *A. niger* (MIC = 78.1 μg/mL), moderately active against *T. mentagrophytes* and *A. fumigatus* (MIC = 156.3 μg/mL), and moderately active against *B. cereus*, *S. epidermidis*, *C. neoformans*, *M. canis*, and *C. albicans* (MIC = 312.5 μg/mL). The antimicrobial activities reported herein complement those reported previously by Sonibare and co-workers who found good activity against *Proteus mirabilis* (agar diffusion assay).[23] It is tenuous to speculate as to what components may be responsible for the antimicrobial activities; there does not seem to be any correlation between the activities of (*E*)-β-caryophyllene, caryophyllene oxide, or phytol and the activities of the essential oil. Incensyl acetate may also be responsible for the antimicrobial activities; incensyl acetate-rich frankincense has shown antimicrobial activities.[29] Synergistic effects likely play a role in the activities.[30–32]

Conclusions

In conclusion, this report complements the previous investigation of Sonibare and co-workers[23] and demonstrates the wide variation in essential oil composition of *V. doniana* leaf essential oil. It also adds to our knowledge of the antifungal properties of this essential oil; *V. doniana* should be considered as an alternative or complementary antifungal therapy option.

Materials and Methods

Plant Material

V. doniana Sweet leaves (2.5 kg) were collected from mature trees in the month of August, 2019, from Ile-Igbon Village, Lagelu, Oyo State, Nigeria at (7°28′59″ N, 4°4′59″ E). Botanical identification was done by Mr. S. A. Odewo at the Herbarium, Forest Research Institute of Nigeria (FRIN),

Table 2. Antibacterial and Antifungal Activities (MIC, μg/mL) of *Vitex doniana* Leaf Essential Oil From Southwestern Nigeria.

Organism	*Vitex doniana* leaf EO	(*E*)-β-Caryophyllene	Caryophyllene Oxide	Phytol	Positive Control[a]
Bacteria					
Bacillus cereus	312.5	312.5	312.5	312.5	1.22
Pseudomonas aeruginosa	625	312.5	312.5	312.5	<19.5
Staphylococcus aureus	1250	312.5	78.1	312.5	0.61
Staphylococcus epidermidis	312.5	312.5	312.5	312.5	< 19.5
Streptococcus pyogenes	625	312.5	625	312.5	< 19.5
Molds					
Aspergillus fumigatus	156.3	156.3	156.3	78.1	< 19.5
Aspergillus niger	78.1	1250	156.3	1250	1.56
Cryptococcus neoformans	312.5	312.5	312.5	312.5	0.78
Microsporum canis	312.5	312.5	312.5	312.5	< 19.5
Serratia marcescens	625	312.5	156.3	312.5	< 19.5
Microsporum gypseum	625	312.5	156.3	156.3	< 19.5
Trichophyton mentagrophytes	156.3	625	156.3	39.1	< 19.5
Trichophyton rubrum	625	312.5	312.5	312.5	< 19.5
Yeast					
Candida albicans	312.5	156.3	312.5	1250	1.56

[a]Gentamicin for bacteria, amphotericin B for fungi.

Jericho, Ibadan, Nigeria, where a voucher specimen (Voucher Number FHI 112549) was deposited. Hydrodistillation of 3 samples of leaves, 450 g each, were carried out using all-glass Clevenger-type apparatus. For each distillation, a ratio of 2:6 *V. doniana* leaves and water were mixed and hydrodistilled for 3 to 4 h until no additional oil was observed to be distilled. The essential oils were combined, dried over anhydrous sodium sulfate to eliminate traces of water, and stored in a sealed vial under refrigeration (4 °C) prior to analysis.

GC-MS Analysis

The leaf essential oil from *V. doniana* was subjected to GC-MS analysis as previously reported [26]: the sample was injected 3 times. Identification of the individual components of the essential oils was prepared by injection of pure samples when available and determined by comparison of the retention index values, which were determined by calibration using a series of *n*-alkanes,[33] in addition to MS fragmentation comparisons with those found in the databases.[19–22]

Antibacterial and Antifungal Assays

The essential oil and 3 different components, (*E*)-β-caryophyllene, caryophyllene oxide, and phytol (Sigma-Aldrich, St. Louis, MO), were screened for antimicrobial activity against bacteria (*Bacillus cereus* [ATCC No. 14579], *Staphylococcus aureus* [ATCC No. 29213], *Staphylococcus epidermidis* [ATCC No. 12228], and *Streptococcus pyogenes* [ATCC No. 19615]); antifungal activity against the molds (*Aspergillus fumigatus* [ATCC No. 96918], *Aspergillus niger* [ATCC No. 16888], *Cryptococcus neoformans* [ATCC No. 32045], *Microsporum canis* [ATCC No. 11621], *Microsporum gypseum* [ATCC No. 24102], *Trichophyton mentagrophytes* [ATCC No. 18748], and *Trichophyton rubrum* [ATCC No. 28188]), along with one yeast (*Candida albicans* [ATCC No. 18804]) using the microbroth dilution technique as previously described.[34,35]

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Data Availability Statement

Data from this investigation are available in the manuscript.

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