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Major Sesquiterpenoids in *Psychotria laui* Leaf Essential oil Exhibit Anti-Inflammatory and Cytotoxic Properties

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Abstract

Objective: This study aims to investigate the chemical constituents and biological properties of the essential oil derived from *Psychotria laui*, a plant recognized for its medicinal applications, particularly in traditional Vietnamese medicine, where it is utilized for the treatment of digestive and inflammatory disorders. **Methods:** Fresh leaves of *Psychotria laui* were gathered in Quangtri, Vietnam, and subjected to hydro-distillation using a Clevenger-type apparatus to obtain the essential oil. The chemical composition of the essential oil was analyzed by gas chromatography-mass spectrometry (GC-MS). The antioxidant potential was measured using the DPPH (2,2-Diphenyl-1-picrylhydrazyl) assay, while the inhibition of nitric oxide production was tested in lipopolysaccharide-stimulated RAW264.7 cells. Cytotoxicity against the SK-LU-1 cell line (human lung adenocarcinoma) was evaluated using the sulforhodamine B (SRB) assay. **Results:** The essential oil from *Psychotria laui* contained 78 distinct constituents. The major compounds identified were spathulenol (10.4%), thujopsan-2-α-ol (9.5%), 7-*epi*-α-eudesmol (6.5%), 14-hydroxy-9-*epi*-(*E*)-caryophyllene (4.4%), *epi*-α-cadinol (3.9%), α-muurolol (3.8%), 1-*epi*-cubenol (3.5%), and δ-cadinene (3.2%). The essential oil showed no antioxidant activity, with an IC_{50} (half-maximal inhibitory concentration) value greater than 500 μg/mL, in contrast to the positive control, L-ascorbic acid, which had an IC_{50} of 7.78 ± 0.39 μg/mL in the DPPH assay. However, its anti-inflammatory activity was evident by its ability to inhibit nitric oxide production, with an IC_{50} of 19.48 ± 1.53 μg/mL in lipopolysaccharide-stimulated RAW264.7 macrophages. Moreover, the essential oil exhibited potent cytotoxicity against the SK-LU-1 cancer cell line, with an IC_{50} of 8.49 ± 0.73 μg/mL in the SRB assay. **Conclusion:** The essential oil from *Psychotria laui* leaves contains numerous bioactive compounds and exhibits significant anti-inflammatory and cytotoxic properties, although it lacks antioxidant activity.

Keywords

Psychotria laui, essential oil, GC-MS, spathulenol, antioxidant, anti-inflammatory, anticancer

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Introduction

Essential oils are intricate blends of plant-derived compounds known for their diverse biological activities, such as cytotoxic, anti-inflammatory, antioxidant, antimicrobial, and antifungal effects.[1] Antioxidants present in essential oils help mitigate oxidative stress by neutralizing harmful radicals, thereby safeguarding cells and promoting overall health.[2] Moreover, secondary metabolites in essential oils, including phenolic compounds, sesquiterpenes, and monoterpenes, are well-recognized for their strong anti-inflammatory properties, mainly by regulating immune response pathways and decreasing inflammation.[3] Chronic inflammation and oxidative stress are interconnected pathogenic processes that contribute to cancer development and progression.[4] Essential oils have remarkable pharmacological and biological characteristics that make them useful for treating a variety of diseases. Thus, research is necessary to find out more concerning the medicinal uses of essential oils.

The genus *Psychotria* stands out among the essential oils of the family Rubiaceae that are derived from natural sources. This genus, which has more than 2000 species, is primarily found in tropical and subtropical regions. various indigenous cultures are familiar with it from its various traditional medical uses.[5,6] Although the chemical compositions of the various *Psychotria* species can differ, common compounds found across many species include flavonoids, terpenoids, alkaloids, and tannins.[5-7] These species display a diverse array of

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biological properties, including antibacterial, anti-inflammatory, antioxidant, and antiparasitic activities.[5,6] Research on *Psychotria* species (*P. asiatica*, *P. eurycarpa*, *P. leiocarpa*, *P. poeppigiana*, and *P. serpens*) has shown distinct compositions, even though the chemical makeup of *Psychotria* species' essential oils has received little to no attention. These studies demonstrate that essential oils contain monoterpenes, sesquiterpenes, and oxygenated sesquiterpenes.[8-12]

In Vietnam, the genus *Psychotria* has approximately twenty-five species, nine of which have been traditionally employed in medicine. *Psychotria laui* has a long-standing history in Vietnamese medicine, where it is utilized for its anti-inflammatory and analgesic effects, especially in the management of joint pain and digestive issues.[13-16] Additional research into the chemistry and biological functions of *P. laui* is essential to enhance our understanding of this species. The objective of this work was to analyze the chemical composition of the essential oil extracted from *P. laui* leaves and evaluate its cytotoxic, anti-inflammatory, and antioxidant potential. This is the first study that, as far as we know, describes the components and biological properties of the essential oil that is extracted from *P. laui* leaves.

Materials and Methods

Hydrodistillation of Essential oil

P. laui leaves were obtained in January 2024 from Quang Tri Province, Vietnam, with geographical coordinates at 16° 38′31.9″N 106°51′36.5″E and co-author Dr Anh Tuan Le (VAST, Vietnam) verified the authenticity of the leaves. The University of Education, Hue University, Vietnam's Faculty of Chemistry has preserved a voucher specimen (PLL-QT-2024). After being freshly picked, the 1.2 kg of leaves were finely chopped and thoroughly cleaned. The essential oil was obtained via hydro-distillation using a 2 L Clevenger apparatus. After the distillation, the essential oil was separated from the aqueous layer and collected with the help of a separating funnel. The process was tuned for maximum oil extraction over 4.0 h. Following extraction, the oil was chilled until additional analysis and dried using Na_2SO_4.[12,17-19]

Analysis of the Volatile Compounds

The volatile components of *P. laui* leaf essential oil were analyzed using a Shimadzu GC-MS QP2010 Plus system from Kyoto, Japan, which integrates mass spectrometry (MS) and gas chromatography (GC) techniques. Following the standard procedures from our earlier research, the chemical components of *P. laui* leaves were identified with accuracy and dependability.[12,17-19] The GC-MS analysis was conducted using a Shimadzu GCMS-QP2010 Plus chromatograph, which was equipped with a fused silica Equity-5 capillary column (30 m × 0.25 mm, 0.25 μm film thickness, Supelco, USA). The temperature program was as follows: the column temperature was initially set to 60 °C for 2 min, then increased to 240 °C at a rate of 3 °C/min, where it was maintained for 10 min, and subsequently raised to 280 °C at a rate of 5 °C/min for an additional 15 min. The injector and interface temperatures were maintained at 280 °C.

DPPH Radical Scavenging Activity

Following established protocols, laboratory conditions were modified as needed, and the capacity of the test sample to scavenge free radicals generated by DPPH was evaluated. The antioxidant activity of *P. laui* essential oil was determined using the DPPH radical scavenging assay. L-Ascorbic acid, a well-established antioxidant, was employed as the positive control, while a solvent or buffer solution devoid of both the essential oil and L-Ascorbic acid served as the negative control.[12,17,19,20]

Anti-Inflammatory Assay

The study investigated the ability of essential oil derived from *P. laui* leaves to inhibit nitric oxide (NO) production in RAW 264.7 cells stimulated with lipopolysaccharide (LPS). The concentration of nitrite, which serves as an indicator of NO presence in the culture media, was quantified using the Griess reaction. Comprehensive methodologies for these assessments can be found in our earlier studies. The anti-inflammatory effects were assessed by measuring the decrease in NO production in LPS-stimulated RAW264.7 macrophage cells. Dexamethasone, recognized for its anti-inflammatory properties, was used as the positive control, while untreated RAW264.7 cells or those treated with the solvent acted as the negative control.[12,17,19,20]

Cytotoxicity Assays

The cytotoxicity of the essential oils was assessed using the sulforhodamine B (SRB) assay on the SK-LU-1 cell line (human lung adenocarcinoma). Detailed methodologies for conducting cytotoxic assays are provided in our prior reports. The cytotoxic effects of the essential oil were tested on the SK-LU-1 cell line, with ellipticine used as the positive control due to its established anticancer properties. The negative control consisted of a solvent or buffer solution devoid of both the essential oil and Ellipticine.[12,20]

Statistical Analysis

All the experiments were triplicated with the data were described as the mean ± standard deviation (SD). The data were analyzed using Microsoft Excel 2020 (Washington, USA).[21]

Results

Phytochemical Analysis

P. laui leaves yielded 0.12% (v/w) of essential oils upon extraction. As presented in Table 1 and Figure 1, a total of 78

Table 1. Chemical Compounds in Psychotria laui Leaf Essential Oil.

No.	RT	RI_E	RI_L	Compound	Concentration (%)
1	7.14	931	932	α-pinene	0.2
2	8.00	957	952	Benzaldehyde	0.1
3	8.61	975	974	β-pinene	0.6
4	10.39	1023	1020	ρ-cymene	0.2
5	10.56	1027	1024	Limonene	0.7
6	11.16	1041	1036	Benzeneacetaldehyde	0.2
7	12.32	1069	1063	*n*-octanol	0.2
8	13.55	1099	1095	Linalool	1.3
9	13.74	1103	1100	*n*-nonanal	0.1
10	16.50	1165	1165	Borneol	0.8
11	17.01	1176	1174	Terpinen-4-ol	0.4
12	17.62	1190	1186	α-terpineol	2.6
13	17.88	1196	1194	Myrtenol	0.2
14	19.17	1225	1224	Hydrocinnamyl alcohol	0.8
15	19.29	1228	1227	Nerol	0.3
16	19.86	1240	1235	(*Z*)-citral	0.2
17	20.46	1254	1249	Geraniol	0.4
18	21.17	1270	1264	(*E*)-citral	0.6
19	21.57	1279	1279	Vitispirane	0.5
20	22.42	1298	1294	Perilla alcohol	0.1
21	22.55	1301	1298	Carvacrol	0.2
22	25.79	1376	1773	α-ylangene	0.2
23	27.64	1420	1417	β-caryophyllene	1.8
24	27.97	1427	1428	(*E*)-α-ionone	0.1
25	28.04	1429	1430	β-copaene	0.2
26	28.45	1439	1437	α-guaiene	0.6
27	29.08	1454	1454	(*E*)-β-farnesene	1.0
28	29.36	1461	1457	β-santalene	0.7
29	29.50	1465	1465	(E)-ethyl cinnamate	0.3
30	30.00	1477	1478	γ-muurolene	1.2
31	30.15	1481	1481	Widdra-2,4(14)-diene	0.3
32	30.39	1486	1487	(*E*)-β-ionone	0.2
33	30.49	1489	1489	α-vetispirene	0.5
34	30.76	1495	1492	δ-selinene	1.0
35	30.97	1500	1500	α-muurolene	0.9
36	31.13	1505	1505	(*E*,*E*)-α-farnesene	0.2
37	31.25	1508	1509	α-bulnesene	0.4
38	31.53	1515	1513	γ-cadinene	2.3
39	31.89	1524	1522	δ-cadinene	3.2
40	32.24	1533	1532	γ-cuprenene	0.1
41	32.47	1539	1537	α-cadinene	0.8
42	32.66	1544	1544	α-calacorene	1.4
43	33.05	1553	1548	Elemol	2.3
44	33.33	1561	1559	Germacrene B	1.0
45	33.46	1564	1561	(*E*)-nerolidol	0.9
46	33.66	1569	1567	Longipinanol	0.9
47	33.74	1571	1570	Dendrolasin	0.6
48	34.09	1580	1577	Spathulenol	10.4
49	34.18	1582	1582	Caryophyllene oxide	1.6
50	34.33	1586	1586	Thujopsan-2-α-ol	9.5
51	34.69	1595	1594	Carotol	1.7
52	34.86	1600	1596	Fokienol	2.1
53	35.06	1604	1602	Ledol	1.1
54	35.38	1611	1608	Humulene epoxide II	1.2
55	35.38	1613	1612	β-biotol	1.0
56	35.49	1617	1618	*epi*-cedrol	1.0
57	35.62	1620	1618	Junenol	0.8
58	35.78	1624	1621	β-cedrene epoxide	2.0

(Continued)

Table 1. Continued.

No.	RT	RI_E	RI_L	Compound	Concentration (%)
59	35.99	1630	1627	1-*epi*-cubenol	3.5
60	36.15	1634	1631	(*E*)-sesquilavandulol	1.6
61	36.31	1638	1638	*epi*-α-cadinol	3.9
62	36.46	1643	1644	α-muurolol	3.8
63	36.66	1648	1646	Agarospirol	1.2
64	36.98	1656	1652	α-cadinol	2.3
65	37.11	1660	1662	7-*epi*-α-eudesmol	6.5
66	37.39	1667	1666	14-hydroxy-(*Z*)-caryophyllene	0.3
67	37.59	1673	1668	14-hydroxy-9-*epi*-(*E*)-caryophyllene	4.4
68	37.72	1676	1675	8,9-epoxide-cadalene	0.5
69	37.91	1682	1679	Khusinol	0.1
70	38.13	1687	1685	α-bisabolol	0.8
71	38.28	1691	1689	(*Z*)-apritone	0.4
72	38.5	1697	1700	Eudesm-7(11)-en-4-ol	0.2
73	39.35	1721	1722	(2*Z*,6*E*)-farnesol	0.5
74	40.87	1764	1759	Benzyl benzoate	0.3
75	47.49	1960	1959	Hexadecanoic acid	1.0
76	48.52	1992	1992	Ethyl hexadecanoate	0.2
77	52.17	2111	2115	7-isoprenyl oxycoumarin	0.9
78	52.90	2135	2132	Linoleic acid	0.2
Total					98.8
Monoterpene hydrocarbons					1.7
Oxygenated monoterpenes					7.6
Sesquiterpene hydrocarbons					18.1
Oxygenated sesquiterpenes					67.1
Non-terpenic compounds					4.3

RT: Retention time, RI_E: Retention indices relative to *n*-alkanes (C_7-C_{40}) on Equity-5 column, RI_L: Retention indices from the Adams book.[22]

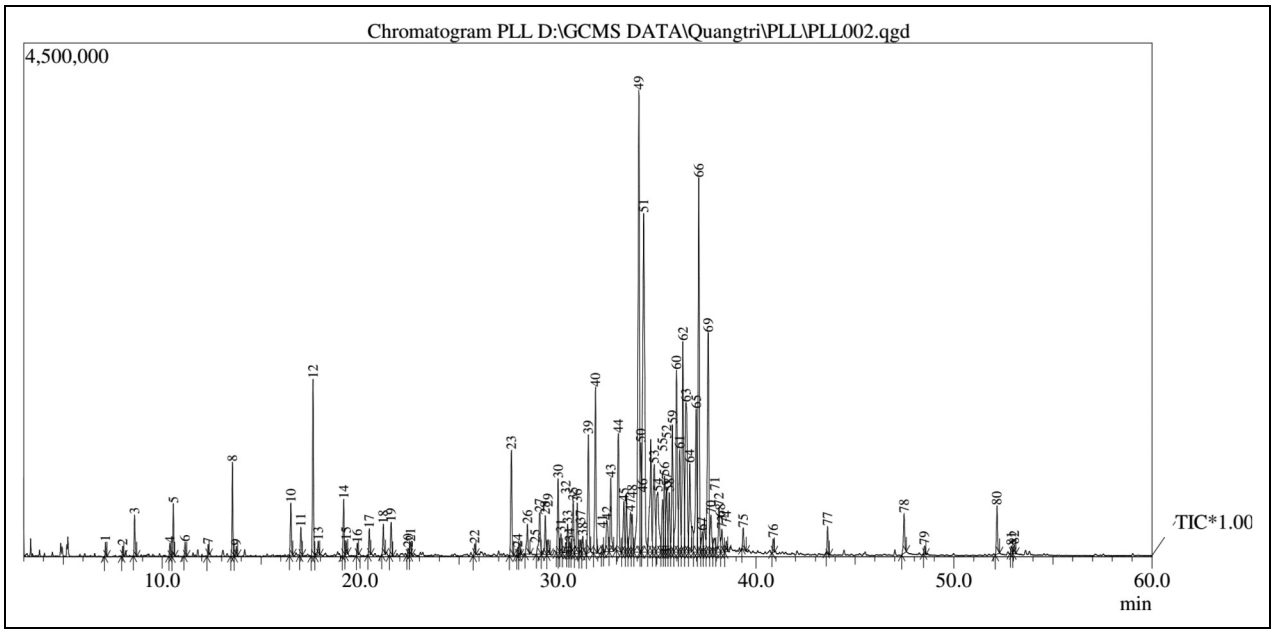

Figure 1. Gc of the essential oil from *Psychotria laui* leaves.

compounds were identified, comprising 98.8% of the total oil content. The main chemical classes found are oxygenated sesquiterpenes (67.1%), sesquiterpene hydrocarbons (18.1%), oxygenated monoterpenes (7.6%), and monoterpene hydrocarbons (1.7%). The main constituents of *P. laui* leaf essential oil, selected based on their concentrations exceeding 3%, include

spathulenol (10.4%), thujopsan-2-α-ol (9.5%), 7-*epi*-α-eudesmol (6.5%), 14-hydroxy-9-*epi*-(*E*)-caryophyllene (4.4%), *epi*-α-cadinol (3.9%), α-muurolol (3.8%), 1-*epi*-cubenol (3.5%), and δ-cadinene (3.2%).

DPPH Radical Scavenging Activity

The antioxidant activity of the essential oil was evaluated using the DPPH assay, as illustrated in Table 2. The essential oil exhibited no significant antioxidant activity, demonstrated by an IC_{50} value exceeding 500 μg/mL.

Anti-Inflammatory Effect

Nitric oxide (NO), a key mediator of inflammatory responses, is overproduced in various clinical disorders. To explore this, we measured the inhibitory effects of *P. laui* essential oil on NO production in LPS-induced RAW264.7 macrophage cells, as presented in Table 3. The essential oil exhibited a significant reduction in NO production, with an IC_{50} value of 19.48 ± 1.53 μg/mL. In comparison, the positive control, dexamethasone, demonstrated an IC_{50} value of 13.71 ± 1.48 μg/mL.

Anti-Cancer Effect

Another study examined the effects of *P. laui* leaf essential oil on the proliferation of the SK-LU-1 cell line utilizing the SRB assay. The findings revealed significant cytotoxicity, with an IC_{50} value of 8.49 ± 0.73 μg/mL (see Table 4).[17,20]

Discussion

This study centered on essential oils derived from the leaves of *P. laui*, utilizing GC-MS analysis to identify the various components of the leaf essential oil. The antioxidant, anti-inflammatory, and anticancer effects were evaluated using DPPH, NO inhibition, and MTT assays, respectively.

The essential oil yield from *P. laui* leaves was 0.12% (v/w). In comparison, *P. poeppigiana* leaves produced a higher yield of 0.4% (w/w).[11] *P. eurycarpa* flowers yielded 0.0629%, which is lower than *P. laui*.[8] The essential oil yield from *P. leiocarpa* leaves was 0.1%, slightly less than that of *P. laui*.[9] *P. serpens*, using its aerial parts, had a yield of 0.05% (w/w), the lowest among the compared species.[10] Lastly, *P. asiatica* leaves yielded 0.08% (v/w), which is also lower than *P. laui*.[12] The observed differences in the chemical profiles between *P. laui* and other *Psychotria* species are based on a comparative analysis of their essential oil compositions. Specifically, we have analyzed the concentration and diversity of key chemical classes and individual compounds. *P. laui* leaves' essential oils contain three main constituents: spathulenol (10.4%), thujopsan-2-α-ol (9.5%), and 7-*epi*-α-eudesmol (6.5%). Prominent research in this field has investigated *P. poeppigiana*'s essential oil and determined that its primary components are bicyclogermacrene (25.21%) and germacrene D (29.38%).[11] Likewise, the principal constituents of *P. eurycarpa*'s floral essential oil were linalool (49.2%) and methyl salicylate (34.1%).[8] Germacrene D (17.6%) and bicyclogermacrene (35.6%) were the two main constituents of *P. leiocarpa* leaf essential oil.[9] Significant components of *P. serpens* were found in another investigation, including linalool (10.0%) and 1-octen-3-ol (23.2%).[10] Major components of *P. asiatica* essential oil were identified to include (*E*)-citral, 10-*epi*-γ-eudesmol, and (*Z*)-citral, with respective values of 20.6, 15.9, and 10.5%.[12] Based on these results, *Psychotria* plants may be useful suppliers of essential oils with a high concentration of oxygenated sesquiterpene hydrocarbons. Variations in the results can be ascribed to pertinent characteristics, collecting times, and geographic disparities, among other things.

There was no noticeable antioxidant action in the essential oil, as indicated by its IC_{50} value of more than 500 μg/mL. Similarly, the DPPH scavenging assay revealed negligible antioxidant activity in *P. asiatica* leaf oil.[12] Conversely, the IC_{50} value of 12.78 ± 1.36 μg/mL for the *P. poeppigiana* leaf essential oil showed considerable antioxidant activity.[11] Furthermore, the *P. serpens* essential oil showed notable ferric ion reduction activity (TEAC value of 46.31 μmol Trolox × g^{-1}) and strong $ABTS^{+}$ radical scavenging activity (IC_{50} value of 0.438 mg/mL). Its IC_{50} value of 1.250 mg/mL indicated that it was not very effective at scavenging DPPH radicals.[10]

The essential oil's IC_{50} value of 19.48 ± 1.53 μg/mL showed that it significantly suppressed NO production. On the other hand, the positive control, dexamethasone, had an IC_{50} value of 13.71 ± 1.48 μg/mL. *P. asiatica* leaf oil has an IC_{50} for NO inhibition in RAW 264.7 cells of 29.08 ± 1.54 μg/mL, according to earlier research employing the MTT test.[12] Research on the potential anti-inflammatory effects of essential oils from the *Psychotria* genus is generally limited. The anti-inflammatory properties of *P. laui* leaf oil are likely due to its primary compounds, including spathulenol, thujopsan-2-α-ol, and 7-*epi*-α-eudesmol. Spathulenol, a sesquiterpene, has been recognized as a significant volatile component in the essential oils of various aromatic species within the Myrtaceae family.[23-27] Examples of these species include *Eugenia calycina*, *E. uniflora*,

Table 2. IC_{50} Values of DPPH Scavenging of Leaf Essential oil of *Psychotria Laui*.

Concentration (μg/mL)	% DPPH scavenging	
	Psychotria laui leaf essential oil	L-Ascorbic acid[a]
500	28.85 ± 0.11	–
100	11.31 ± 1.20	91.79 ± 2.26
20	1.69 ± 0.87	72.51 ± 1.13
4	-0.31 ± 0.33	32.88 ± 1.04
0.8	–	14.27 ± 0.91
IC_{50}^{b}	>500	7.78 ± 0.39

[a]Positive control.
[b]Scavenging Concentration at 50% – concentration that neutralizes 50% of DPPH free radicals.

Table 3. *in vitro* Anti-Inflammatory Activity of Leaf Essential oil of *Psychotria Laui.*

	Psychotria laui leaf essential oil		Dexamethasone[a]	
Concentration (μg/mL)	NO inhibition rate (%)	Viability rate (%)	NO inhibition rate (%)	Viability rate (%)
100	94.82 ± 2.49	10.04 ± 1.10	90.58 ± 2.12	95.18 ± 2.44
20	51.02 ± 1.88	81.50 ± 1.38	54.23 ± 1.17	98.56 ± 1.93
4	22.04 ± 1.37	–	40.64 ± 1.08	–
0.8	12.94 ± 0.63	–	32.59 ± 1.13	–
IC_{50}^{b}	19.48 ± 1.53		13.71 ± 1.48	

[a]Positive control.
[b]Concentration that inhibits 50% of cell growth.

Table 4. Cytotoxicity of the Essential oil from *Psychotria laui* Leaves Against Human Cancer Cell Lines.

Concentration (μg/mL)	SK-LU-1	
	Psychotria laui leaf essential oil (% inhibition)	Ellipticine[a] (% inhibition)
100	98.47 ± 1.10	89.55 ± 1.32
20	75.39 ± 1.52	84.59 ± 1.84
4	26.70 ± 1.72	48.45 ± 1.12
0.8	15.29 ± 1.08	21.69 ± 0.86
IC_{50}^{b}	8.49 ± 0.73	0.42 ± 0.02

[a]Positive control; SK-LU-1: Human lung adenocarcinoma cell line.
[b]The half-maximal inhibitory concentration.

Psidium cattleianum, P. guineense, and *P. guajava*.[23-27] Studies on spathulenol-rich essential oils have demonstrated notable anti-inflammatory properties.[27-29] Another important molecule is 7-*epi*-α-eudesmol, a sesquiterpenoid of the eudesmane class. The eudesmane sesquiterpenoids found in the fruits of *Alpinia oxyphylla* are a diversified group with notable anti-inflammatory characteristics. The ability of these compounds to decrease the generation of NO in LPS-stimulated BV-2 cells has been assessed; IC_{50} values range from 21.63 to 60.70 μM, indicating moderate to substantial NO suppression. Additionally, these compounds partially inhibited the production of TNF-α and IL-6 in LPS-stimulated BV-2 cells. The mRNA expressions of TNF-α, IL-6, COX-2, and *i*NOS were significantly reduced, while the protein expressions of COX-2 and *i*NOS were also downregulated.[30,31]

The results showed considerable cytotoxicity with an IC_{50} value of 8.49 ± 0.73 μg/mL (Table 4).[17,20] However, in a comparable study evaluating *P. asiatica* leaf essential oil, SK-LU-1 cells demonstrated a higher IC_{50} value of 39.75 ± 1.79 μg/mL.[12] These findings draw attention to the small but growing corpus of research regarding the potential anti-cancer properties of essential oils sourced from the *Psychotria* genus. The leaf extract of *Dasymaschalon dasymaschalum*, spathulenol, has been shown to have strong cytotoxic effects on human lung cancer cell lines (NCI-H187), with an IC_{50} value of 6.6 μg/mL.[32] Conversely, spathulenol isolated from the aerial parts of *Glycosmis parviflora* demonstrated cytotoxicity against human lung cancer cell lines, SK-LU-1, with an IC_{50} value of 41.86 ± 2.58 μg/mL.[33] Spathulenol also exhibits cytotoxicity against human gastric adenocarcinoma (AGS), murine melanoma (B16-F10), human hepatoblastoma (HepG2), human leukemia (HL-60 and K562), and human ovarian cancer (OVCAR-3).[27,34,35] Several studies have demonstrated the moderate to strong cytotoxic effects of essential oils (EOs) enriched in spathulenol against a variety of cancer cell lines, such as colorectal cancer (CACO-2), human hepatocellular carcinoma (Hep-G2), human ovarian carcinoma (HeLa), human hepatocellular carcinoma (Bel-7402), ovarian carcinoma (A-549, MCF-7, and A2780).[36-40] In addition to its cytotoxic effects, spathulenol has been linked to several biological activities, such as the suppression of the human ABCB1 efflux pump and its anticancer, immunomodulatory, and antibacterial qualities.[40,41]

Significant concentrations of spathulenol, thujopsan-2-α-ol, and 7-*epi*-α-eudesmol have been identified in the essential oil extracted from *P. laui* leaves, suggesting potential applications in cytotoxic and anti-inflammatory contexts. Notably, the presence of thujopsan-2-α-ol points to possible therapeutic benefits, although its biological functions have not been documented previously. Our study assessed the antioxidant, anti-inflammatory, and anticancer properties of *P. laui* essential oil. It is important to highlight several limitations: Firstly, the essential oil showed no antioxidant activity in our assays, suggesting a potential ineffectiveness in scavenging free radicals or providing protection against oxidative stress. Secondly, while the oil demonstrated significant anti-inflammatory effects, as shown by a marked reduction in nitric oxide production in LPS-induced RAW264.7 macrophage cells, this finding is derived from *in vitro* models, which may not fully capture the complexity of *in vivo* inflammatory responses. Finally, the essential oil displayed potent cytotoxic effects in the SK-LU-1 cell line. However, this result is limited to a single cancer cell line, necessitating further research to explore its efficacy against a broader range of cancer types and in animal models. Addressing these limitations in future studies will be crucial for a more comprehensive understanding of the biological activities of *P. laui* essential oil.

Conclusion

Our work is the first of its kind since it provides new information on the biological characteristics and chemical composition of the essential oil derived from *P. laui* leaves. A total of

seventy-eight distinct compounds were identified in the essential oil extracted from *P. laui* leaves, with the three primary components being spathulenol (10.4%), thujopsan-2-α-ol (9.5%), and 7-*epi*-α-eudesmol (6.5%). Minimal antioxidant activity was found in the DPPH experiment, with an IC_{50} value higher than 500 μg/mL. The reduction in NO production in LPS-induced RAW264.7 macrophage cells was used to measure the anti-inflammatory potential. A substantial difference was seen in the results ($IC_{50} = 19.48 \pm 1.53$ μg/mL). Moreover, the SK-LU-1 cell line had strong cytotoxic effects, with an IC_{50} value of 8.49 ± 0.73 μg/mL. Further investigation into *P. laui* leaf essential oil is warranted due to its promising anti-inflammatory and anticancer characteristics.

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Authors Contributions

Tien Dong Tran and Ty Viet Pham conceived and designed research. Tien Dong Tran, Anh Tuan Le, Thang Quoc Le, and Ty Viet Pham conducted experiments and analyze data. Tien Dong Tran and Ty Viet Pham wrote the manuscript. All authors read and approved the manuscript.

Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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Ethical Approval

Not applicable, because this article does not contain any studies with human or animal subjects.

Statement of Human and Animal Rights

Not applicable, because this article does not contain any guidelines followed for performing experimental procedures.

Informed Consent

Not applicable, because this article does not contain any studies with human or animal subjects.

Trial Registration

Not applicable because this article does not contain any clinical trials.

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