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Chemical Composition By Gc-Ms And ^{13}C Nmr Analyses Of The Constituents Of The Essential Oil From The Leaves Of *Piper Guineense* Schumach. & Thonn. From Côte D'ivoire: Predominance Of Hydrogenated Azulene And B-Caryophyllene

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Abstract
The essential oil extracted by hydrodistillation from the leaves of Piper guineense Schumach. & Thonn. (Côte d'Ivoire) showed a yield of 2.00 ± 0.01% (w/w). Combined analysis using gas chromatography–mass spectrometry (GC-MS) and ^{13}C nuclear magnetic resonance (^{13}C NMR) spectroscopy enabled the identification of 41 volatile compounds, classified into five groups: sesquiterpenes (92.46%), diterpenes (4.53%), monoterpenes (2.74%), linear alcohols (0.25%), and linear hydrocarbons (0.29%). The major constituents were hydrogenated azulene (20.54%), β-caryophyllene (19.54%), 1-(1-methylethyl)-4, 7-dimethyl-1,2,3,5,6, 8a-hexahydronaphthalene (7.95%), β-farnesene (5.27%), decahydro-1,1,4,7-tetramethyl-1H cycloprop [e] azulen-4-ol (5.02%), phytol (4.07%), 1-hydroxy-1, 6-dimethyl-4-(1-methylethyl) octahydronaphthalene (3.98%), and caryophyllene oxide (3.68%). In particular, sesquiterpenes overwhelmingly dominated the volatile profile, with a striking predominance of hydrogenated azulene and β-caryophyllene.

Keywords : Piper guineense*; Essential oil; GC-MS; ^{13}C NMR; Sesquiterpenes; β-Caryophyllene; Hydrogenated azulene*

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I. Introduction

Piper guineense Schumach. & Thonn., commonly known as West African pepper or Uziza, belongs to the family *Piperaceae* and represents a plant species of major importance in West Africa, both for its culinary uses and medicinal properties. In Côte d'Ivoire, this plant holds a privileged position in traditional medicine, where it is renowned for its antioxidant, anti-inflammatory, and antimicrobial activities (**Kambiré *et al.,* 2019**; **Juliani *et al.,* 2013**).

The organs of *P. guineense* (fruits, seeds, and leaves) contain a wide diversity of specialized metabolites, including alkaloids, flavonoids, phenolic compounds, and essential oils, which confer on the species a broad range of pharmacological activities (**Ogunmefun *et al.,* 2017; Carsono *et al.,* 2022**). These biomolecules play a key role in the prevention and management of various disorders, including microbial infections, chronic inflammatory diseases, and metabolic pathologies such as diabetes and dyslipidemia (**Akinloye *et al.,* 2020**).

In addition to its therapeutic importance, *P. guineense* occupies a notable place in West African gastronomy, where its leaves and seeds—whether fresh or dried—are used to enhance the flavor of dishes while providing digestive and tonic benefits. This duality between culinary and medicinal functions explains the growing scientific interest in the study of its bioactive constituents. Recent studies have particularly focused on the chemical characterization of its phenolic compounds, the evaluation of its antioxidant potential, and the

analysis of its volatile fractions by gas chromatography–mass spectrometry (GC-MS), providing deeper insight into its therapeutic potential (**Sulaimon *et al.*, 2020**; **Madu *et al.*, 2023**).

The ethnomedicinal uses of *P. guineense* are supported by several scientific studies. **Akinloye *et al.* (2020)** demonstrated that extracts from the plant's leaves and seeds exert significant anti-inflammatory activity in animal models, thus validating traditional claims. Similarly, **Carsono *et al.* (2022)** and **Evuen *et al.* (2022)** reported the use of *P. guineense* infusions and decoctions in the treatment of gastrointestinal disorders, respiratory infections, rheumatism, and venereal diseases. More recently, investigations have revealed potential immunomodulatory, neuroprotective, and anticancer effects, highlighting promising perspectives for its integration into modern pharmacology as well as the cosmetic and agri-food industries (**Ademuyiwa *et al.*, 2023**; **Mgbeahuruike *et al.*, 2019**).

In this context, an in-depth characterization of the essential oil from *P. guineense* leaves—particularly using ^{13}C NMR spectroscopy—is essential to better understand the chemical composition of this plant resource and to valorize its major constituents such as β-caryophyllene and hydrogenated azulene. Such an approach not only contributes to advancing phytochemical knowledge but also paves the way for the development of therapeutic formulations and high-value natural products.

II. Materials And Methods

Plant Material

The plant material used in this study consisted of the leaves of *Piper guineense*, collected in the Daloa region (Côte d'Ivoire) in March 2023. After harvesting, the leaves were kept at room temperature for 24 hours before being used for extraction.

Extraction of the Essential Oil (EO) by Hydrodistillation

Approximately 1.5 L of water was poured into the distillation flask. A measured mass of *P. guineense* leaves was weighed and placed in the flask, which was then sealed and brought to a boil. The extraction was continued for 3 hours from the appearance of the first drop of essential oil (EO). This operation was repeated several times to optimize the yield.

The extracted essential oils were weighed, stored in amber glass bottles, and kept in a freezer at a temperature close to 0 °C.

The yield (Y) for each essential oil sample was calculated according to the following equation:

$$\text{Rdt (\%)} = \frac{\text{Mass of EO obtained (g)}}{\text{Mass of dried plant material (g)}} \times 100$$

Characterization by GC-MS and ^{13}C NMR

For the analysis of the essential oil extracted from *Piper guineense* leaves, two techniques were employed: carbon-13 nuclear magnetic resonance (^{13}C NMR) spectroscopy and gas chromatography (GC) coupled with mass spectrometry (MS). The chromatogram of the essential oil, as well as the mass and ^{13}C NMR spectra of each compound, were recorded using a Bruker instrument (Bruker BioSpin AG). The elution solvent used was deuterated chloroform ($CDCl_3$), and chemical shifts (δ, in ppm) were referenced to tetramethylsilane (TMS) as the internal standard. The instrument was equipped with a 5–10 mm probe operating at 100.623 MHz for carbon-13. The ^{13}C NMR spectra were recorded under the following conditions: 5 mm probe, 45° pulse angle, acquisition time of 2.73 s, acquisition of 64 K data points with a spectral width (SW) of 25,000 Hz (250 ppm), and a digital resolution of 0.183 Hz/point. For the analysis, 70 mg of essential oil were dissolved in 5 mL of $CDCl_3$, and the number of accumulations ranged from 2,000 to 5,000. Decoupling was performed using pulsed field Composite Phase Decoupling (CPD). Before the Fourier transform, the free induction decay (FID) data were multiplied by an exponential function with LB = 1.0 Hz.

Chemical Composition of the Essential Oil from *Piper guineense* Leaves

The essential oil extracted from the leaves of *Piper guineense* was analyzed by gas chromatography coupled with mass spectrometry (GC-MS). The chromatogram of the oil is shown in Figure 1. It reveals the presence of several peaks corresponding to different volatile compounds detected between 3 and 25 minutes.

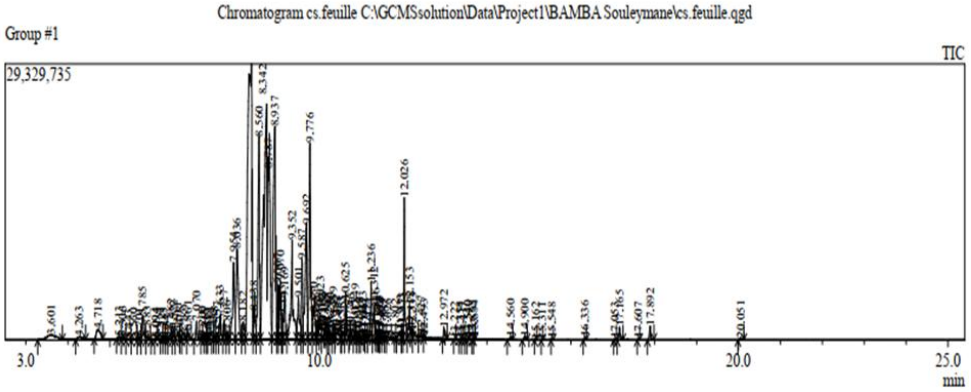

Figure 1: GC-MS chromatogram of the essential oil extracted from the leaves of *P. guineense*.

Combined analyses by gas chromatography coupled with mass spectrometry (GC-MS) and carbon-13 nuclear magnetic resonance (^{13}C NMR) spectroscopy enabled the identification and characterization of 41 volatile constituents in the essential oil extracted from the leaves of *P. guineense*.

The detailed results from the GC-MS analysis, including the names of the compounds, their retention times, and their relative abundances (%), are presented in Table 1.

Table 1: Chemical composition of the essential oil from the leaves of *Piper guineense*.

No.	Retention Time (min)	Compound Name	Relative Content (%)
1	4.263	α-Pinene	0.24
2	4.718	β-Pinene	0.65
3	5.443	Camphane	0.02
4	5.785	Linalool	0.86
5	6.611	(S)-1-(Hydroxymethyl)-1,1-dimethyl-4-methylcyclohex-3-ene	0.26
6	6.678	(2Z)-3-Pentylpent-2,4-dien-1-ol	0.02
7	6.789	4-Methyl-1,1-dimethylcyclohex-3-ene-1-carbaldehyde	0.11
8	6.861	trans-β-Terpinyl pentanoate	0.17
9	7.070	2-Decenal	0.38
10	7.200	Epoxy-α-terpinyl acetate	0.03
11	7.459	Geranylgeraniol	0.01
12	7.488	(2E,4E)-Deca-2,4-dienal	0.02
13	7.633	(3R)-1-(1-Methylethyl)-3-(1-methylethenyl)-4-methylcyclohex-1-en-4-ylethene	0.59
14	7.727	α-Cubebene	0.38
15	7.806	1,1,6-Trimethyldihydronaphthalene	0.20
16	7.954	Copaene	2.71
17	8.036	(1α,2β,4β)-1-Ethenyl-1-methyl-2,4-bis(1-methylethenyl)cyclohexane	3.47
18	8.182	(±)-1-(1-Methylethyl)-4,7-dimethyloctahydronaphthalene	0.58
19	8.342	β-Caryophyllene	19.54
20	8.438	Decahydro-1,1,7-trimethyl-4-methylene-1H-cycloprop[e]azulene	0.97
21	8.560	β-Farnesene	5.27
22	8.787	Hydrogenated azulene	20.54
23	8.937	1-(1-Methylethyl)-4,7-dimethyl-1,2,3,5,6,8a-hexahydronaphthalene	7.95
24	9.007	Di-epimer of α-Cedrene	0.66
25	9.070	α-Calacorene	1.85
26	9.113	Farnesol	0.35
27	9.169	4,8,12-Trimethyl-3,7,11-tridecatrienenitrile	1.26
28	9.352	Caryophyllene oxide	3.68
29	9.501	2,5,9-Trimethylcycloundeca-4,8-dienone	1.40
30	9.587	Cubenol	1.54
31	9.692	1-Hydroxy-1,6-dimethyl-4-(1-methylethyl)octahydronaphthalene	3.98
32	9.776	Decahydro-1,1,4,7-tetramethyl-1H-cycloprop[e]azulen-4-ol	5.02
33	9.860	Decahydro-1,1,7-trimethyl-1H-cycloprop[e]azulene-4-methylene	1.35
34	10.023	Decahydro-1,4a-dimethyl-7-(1-methylethylidene)-1-naphthol	0.97
35	10.073	Tetrahydro-1,3,6,8-tetraethenyldibenzo[a,h]cyclotetradecene	0.60
36	10.849	3,7,11,15-Tetramethylhexadec-2-en-1-ol	0.21
37	11.236	1-(α-Ethenyldecahydro-α,5,5,8a-tetramethyl-2-methylene)naphthalenepropanol	1.10
38	11.346	Thunbergol	0.15
39	12.026	Phytol	4.07
40	12.972	n-Tetracosan-1-ol	0.25
41	17.892	Tetratetracontane	0.29

Among the identified compounds, eight major molecules were selected as representative of the chemical composition of this essential oil. These include hydrogenated azulene (20.54%), β-caryophyllene (19.54%), 1-(1-methylethyl)-4,7-dimethyl-1,2,3,5,6,8a-hexahydronaphthalene (7.95%), β-farnesene (5.27%), decahydro-1,1,4,7-tetramethyl-1H-cycloprop[e]azulen-4-ol (5.02%), phytol (4.07%), 1-hydroxy-1, 6-dimethyl-4-(1-methylethyl) octahydronaphthalene (3.98%), and caryophyllene oxide (3.68%).

The ^{13}C NMR spectra obtained for these compounds exhibit characteristic signals confirming their respective molecular structures:

Hydrogenated azulene (20.54%) shows signals at δ 138.7 and 127.9 ppm corresponding to residual sp^2 carbons, and at δ 48.3, 44.2, and 40.1 ppm assigned to quaternary cyclic carbons. Shifts at δ 34.8, 29.6, and 27.5 ppm are characteristic of saturated CH/CH_2 carbons, while those at δ 22.4 and 18.5 ppm correspond to methyl (CH_3) carbons.

β-Caryophyllene (19.54%) exhibits signals at δ 155.0, 139.1, 133.7, and 124.2 ppm corresponding to olefinic carbons (C=C), as well as at δ 48.5, 46.8, 43.2, and 41.5 ppm assigned to quaternary and bridgehead carbons. Signals at δ 36.9, 34.5, 32.7, and 28.5 ppm indicate the presence of cyclic CH and CH_2 groups, while those at δ 22.1, 19.8, and 15.9 ppm are typical of methyl (CH_3) groups.

1-(1-Methylethyl)-4,7-dimethyl-1,2,3,5,6,8a-hexahydronaphthalene (7.95%) displays signals at δ 47.1 and 42.6 ppm assigned to quaternary/bridgehead carbons of the decahydronaphthalene core. Signals at δ 41.2, 38.3, 36.8, 34.6, 32.9, 31.5, 28.1, 27.2, and 25.7 ppm correspond to cyclic CH and CH_2 groups (including the isopropyl CH around 33 ppm), while those at δ 23.2, 22.8, 20.5, and 18.9 ppm are characteristic of methyl (CH_3) groups (two i-Pr CH_3 around 22–23 ppm and two ring CH_3 around 19–21 ppm).

β-Farnesene (5.27%) is characterized by resonances at δ 136.8, 131.4, 124.7, and 123.2 ppm, typical of olefinic carbons (C=C). Signals at δ 38.9, 32.7, and 29.6 ppm indicate allylic CH_2 groups, while those at δ 27.1 and 25.8 ppm are associated with the aliphatic chain. Signals at δ 17.8 and 16.1 ppm correspond to vinyl methyl groups.

Decahydro-1,1,4,7-tetramethyl-1H-cycloprop[e]azulen-4-ol (5.02%) presents a distinct signal at δ 72.5 ppm assigned to the carbon linked to the hydroxyl function (C–O). Resonances at δ 48.7, 43.6, and 40.2 ppm correspond to quaternary carbons, while those at δ 36.4, 32.1, and 29.8 ppm are characteristic of cyclic CH and CH_2 carbons. Signals at δ 25.6 and 22.7 ppm are assigned to CH_2 groups, and those at δ 19.4 and 17.2 ppm correspond to methyl (CH_3) groups.

Phytol (4.07%) exhibits signals at δ 131.9 and 124.1 ppm indicating the presence of double bonds (C=C), along with a signal at δ 63.2 ppm corresponding to the carbon of the alcohol group (CH_2–OH). Resonances at δ 39.5 and 37.1 ppm are associated with methine carbons, those at δ 32.8, 31.4, and 29.7 ppm with CH_2 groups of the aliphatic chain, while the signals at δ 25.9 and 22.6 ppm reflect other CH_2 groups. Finally, the signals at δ 19.8 and 16.2 ppm correspond to terminal methyl carbons.

1-Hydroxy-1,6-dimethyl-4-(1-methylethyl)octahydronaphthalene (3.98%) shows a prominent signal at δ 71.5 ppm corresponding to the tertiary carbon bearing the hydroxyl group (C–O), along with signals at δ 46.9 and 41.8 ppm assigned to quaternary/bridgehead carbons. Signals at δ 40.3, 37.2, 35.4, 33.0, 31.1, 29.4, 27.9, and 26.3 ppm indicate cyclic CH and CH_2 carbons (including the i-Pr CH near 32–34 ppm). Finally, signals at δ 23.1, 22.7, 20.2, and 18.6 ppm are characteristic of methyl (CH_3) carbons, with the two i-Pr CH_3 groups near 22–23 ppm.

Caryophyllene oxide (3.68%) shows signals at δ 150.3 and 124.6 ppm corresponding to olefinic carbons (C=C), and at δ 69.1 and 58.7 ppm attributed to carbons bound to oxygen in the epoxide ring. Resonances at δ 48.2, 44.6, 41.2, and 38.5 ppm are assigned to bridgehead carbons, those at δ 33.7, 30.4, and 28.1 ppm to cyclic CH_2 groups, and finally, signals at δ 22.0 and 16.7 ppm correspond to methyl (CH_3) groups.

The major identified compounds are illustrated in Figure 2.

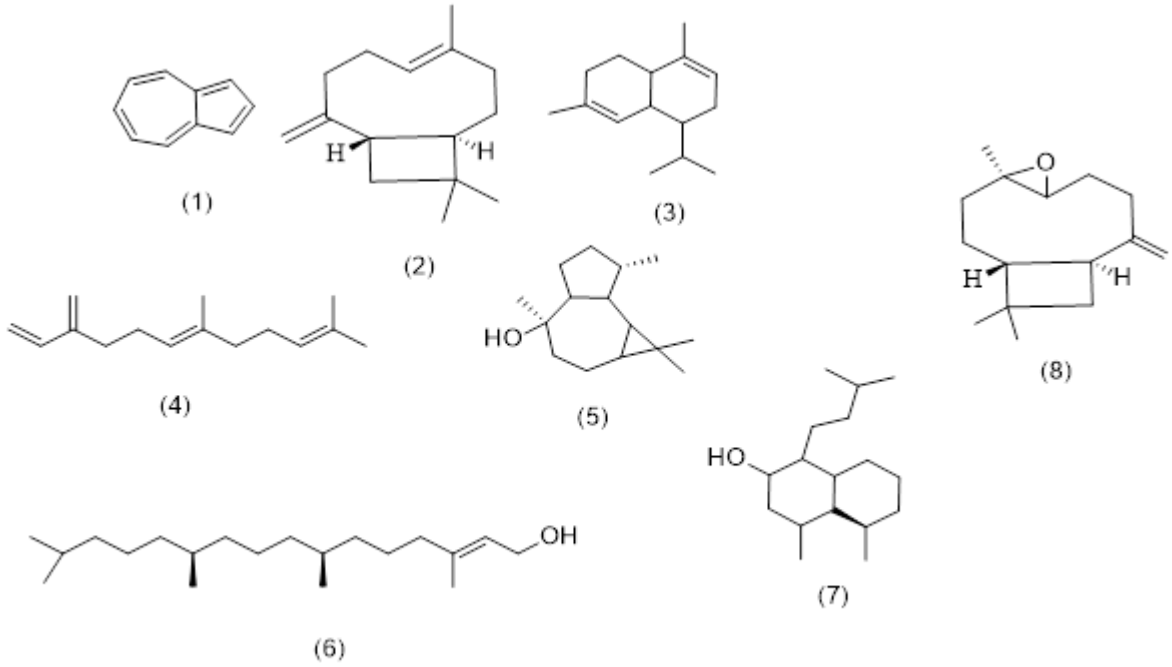

Figure 2: Main compounds identified in the essential oil from the leaves of *Piper guineense* by GC-MS and confirmed by ¹³C NMR. (1) Hydrogenated azulene, (2) β-Caryophyllene, (3) 1-(1-Methylethyl)-4,7-dimethyl-1,2,3,5,6,8a-hexahydronaphthalene, (4) β-Farnesene, (5) Decahydro-1,1,4,7-tetramethyl-1H-cycloprop[e]azulen-4-ol, (6) Phytol, (7) 1-Hydroxy-1,6-dimethyl-4-(1-methylethyl)octahydronaphthalene, (8) Caryophyllene oxide.

To better understand the structural diversity and chemical distribution of the identified compounds, the constituents of the essential oil from *Piper guineense* leaves were classified according to their main chemical families. This classification highlights the predominance of sesquiterpenes (C_{15}), followed by diterpenes (C_{20}) and monoterpenes (C_{10}), while linear alcohols and linear hydrocarbons are present in much lower proportions. The distribution of the identified compounds by chemical family and their respective total yields (%) are presented in Table 2.

Table 2: Distribution of the compounds identified in the essential oil from the leaves of *Piper guineense* according to their chemical families.

Chemical Family	Compounds	Total Yield (%)
Monoterpenes (C_{10})	α-Pinene, β-Pinene, Camphane, Linalool, (S)-1-(Hydroxymethyl)-1,1-dimethyl-4-methylcyclohex-3-ene, (2Z)-3-Pentylpent-2,4-dien-1-ol, 4-Methyl-1,1-dimethylcyclohex-3-ene-1-carbaldehyde, trans-β-Terpinyl pentanoate, 2-Decenal, Epoxy-α-terpinyl acetate	2.74
Sesquiterpenes (C_{15})	Geranylgeraniol, (2E,4E)-Deca-2,4-dienal, (3R)-1-(1-Methylethyl)-3-(1-methylethenyl)-4-methylcyclohex-1-en-4-ylethene, α-Cubebene, 1,1,6-Trimethyldihydronaphthalene, Copaene, (1α,2β,4β)-1-Ethenyl-1-methyl-2,4-bis(1-methylethenyl)cyclohexane, (±)-1-(1-Methylethyl)-4,7-dimethyloctahydronaphthalene, β-Caryophyllene, Decahydro-1,1,7-trimethyl-4-methylene-1H-cycloprop[e]azulene, β-Farnesene, Hydrogenated azulene, 1-(1-Methylethyl)-4,7-dimethyl-1,2,3,5,6,8a-hexahydronaphthalene, Di-epimer of α-Cedrene, α-Calacorene, Farnesol, 4,8,12-Trimethyl-3,7,11-tridecatrienenitrile, Caryophyllene oxide, 2,5,9-Trimethylcycloundeca-4,8-dienone, Cubenol, 1-Hydroxy-1,6-dimethyl-4-(1-methylethyl)octahydronaphthalene, Decahydro-1,1,4,7-tetramethyl-1H-cycloprop[e]azulen-4-ol, Decahydro-1,1,7-trimethyl-1H-cycloprop[e]azulene-4-methylene	92.46
Diterpenes (C_{20})	3,7,11,15-Tetramethylhexadec-2-en-1-ol, 1-(α-Ethenyldecahydro-α,5,5,8a-tetramethyl-2-methylene)naphthalenepropanol, Thunbergol, Phytol	4.53
Linear Alcohols	n-Tetracosan-1-ol	0.25
Linear Hydrocarbons	Tetratetracontane	0.29

III. Discussion

The essential oil yield (2.00%) observed in this study falls within the range generally reported for *Piper guineense* leaves, with values between 0.5% and 3% described in the literature (**Sulaimon *et al.,* 2020**; **Madu *et al.,* 2023**; **Tchoumbougnang *et al.,* 2009**).

The overwhelming predominance of sesquiterpenes (92.46%) indicates that the volatile fraction of *P. guineense* consists mainly of moderately volatile, high-molecular-weight compounds, a feature already observed in other *Piper* essential oils (**Carsono *et al.,* 2022**).

The relatively low contributions of diterpenes (4.53%) and monoterpenes (2.74%) may be attributed either to an intrinsically reduced biosynthesis of these compounds in the leaf tissues of the studied population or to partial thermal degradation during hydrodistillation, since monoterpenes are more sensitive to extraction conditions (**Sulaimon *et al.,* 2020**). The minor presence of linear alcohols and hydrocarbons (<0.3%) corresponds to the typical levels reported for many essential oils, where such compounds are generally considered accessory constituents (**Burt, 2004**).

The identification of hydrogenated azulene (20.54%) and β-caryophyllene (19.54%) as the major compounds gives this oil particular interest. β-Caryophyllene is well documented in essential oils of *Piper* species and other aromatic genera such as *Rosmarinus* and *Syzygium* (**Gertsch *et al.,* 2008**; **Sharma *et al.,* 2021**). The co-occurrence of β-farnesene, caryophyllene oxide, and various hydrogenated naphthalenes reflects a diversified terpenoid metabolism, as also reported in other *Piper* species (**Ogunmefun *et al.,* 2017**).

This study also confirms the usefulness of combining ^{13}C NMR and GC-MS for essential oil analysis. Indeed, the integration of GC-MS and ^{13}C NMR data provides a robust approach for structural identification, particularly for sesquiterpenes whose isomers may exhibit similar mass spectra (**Adams, 2007**; **Mitaine-Offer *et al.,* 2002**; **Santos *et al.,* 2002**). The accurate assignment of ^{13}C signals to characteristic carbons—especially quaternary and substituted carbons—further reinforces the reliability of compound identification (**Sharma *et al.,* 2021**).

However, the detection of 41 constituents suggests that some trace or thermolabile compounds may have escaped identification, a limitation also noted in other studies on sesquiterpene-rich essential oils (**Sulaimon *et al.,* 2020**).

Among the identified compounds, two sesquiterpenes stand out: hydrogenated azulene (20.54%) and β-caryophyllene (19.54%). Azulene and its derivatives are widely known for their anti-inflammatory, soothing, wound-healing, antioxidant, and antiallergic properties, explaining their frequent use in cosmetics and dermatology (**Slon *et al.,* 2024**; **Bakun *et al.,* 2021**). Studies have demonstrated their ability to inhibit lipid peroxidation and reduce inflammation in various experimental models (**Kourounakis *et al.,* 1997**). Moreover, certain azulenes have shown phototoxic and genotoxic effects under irradiation, highlighting the need for thorough safety evaluation prior to topical use (**Chiang *et al.,* 2010**; **Struwe *et al.,* 2011**). β-Caryophyllene, on the other hand, is one of the most extensively studied sesquiterpenes. Its high proportion in *P. guineense* essential oil has significant pharmacological implications. This bicyclic sesquiterpene is a selective agonist of the cannabinoid receptor CB_2, with no affinity for CB_1, which explains its lack of psychotropic effects (**Gertsch *et al.,* 2008**). Numerous *in vitro* and *in vivo* studies have demonstrated its anti-inflammatory, antioxidant, immunomodulatory, and gastroprotective activities (**Sharma *et al.,* 2021**; **Fidyt *et al.,* 2016**). For instance, β-caryophyllene inhibits the expression of pro-inflammatory cytokines (TNF-α, IL-1β) and modulates MAPK/ERK signaling pathways, contributing to the regulation of immune responses (**Sharma *et al.,* 2021**). The coexistence of these two major metabolites may result in synergistic effects, enhancing the overall biological activity of the essential oil, as reported in other sesquiterpene-rich plant matrices (**Bassolé *et al.,* 2012**; **Bakkali *et al.,* 2008**; **Wagner *et al.,* 2003**).

The presence of β-farnesene (5.27%) is also noteworthy. This acyclic sesquiterpene is well known as an aphid alarm pheromone and may play an ecological role in protecting the plant against herbivores (**Beale *et al.,* 2006**). Moreover, several studies have demonstrated its insect-repellent properties and potential applications in crop protection (**Pickett & Khan, 2016**). Copaene (2.71%) and other oxygenated sesquiterpenes such as caryophyllene oxide (3.68%) and cubenol (1.54%) further enrich this profile; these molecules are associated with documented antimicrobial and antioxidant activities, reinforcing the overall therapeutic potential of the oil (**Baser & Buchbauer, 2015**).

The predominance of sesquiterpenes gives this oil a distinct chemical profile, typically referred to as a *sesquiterpenic chemotype*. Such a composition contrasts with that of many common essential oils dominated by monoterpenes (e.g., lavender or eucalyptus oils) and reflects a specific metabolic orientation toward the mevalonate (MVA) pathway, which generates C_{15} precursors (**Cheng *et al.,* 2007**). Although diterpenes were present in smaller proportions (4.53%), they also make a meaningful contribution. Phytol (4.07%), a diterpene alcohol derived from the chlorophyll side chain, possesses antioxidant and antimicrobial properties and is used as an additive in pharmaceutical and cosmetic formulations (**Santos *et al.,* 2013**). Its anti-inflammatory and

antinociceptive activities have also been demonstrated in several experimental models, suggesting that it may act synergistically with sesquiterpenes to enhance the biological activity of the oil (**De Moraes *et al.,* 2014**).

Conversely, monoterpenes account for only a minor fraction (2.74%). α-Pinene and β-Pinene, identified at 0.24% and 0.65% respectively, are known for their antimicrobial and expectorant properties, but their low abundance limits their pharmacological contribution in this extract (**Salehi *et al.,* 2019**). The near absence of monoterpenes also suggests that the studied oil is less volatile and more stable than C_{10}-dominated oils, which tend to oxidize rapidly upon exposure to air and light (**Adams, 2007**). This enhanced stability represents an advantage for industrial and cosmetic applications requiring long shelf lives.

From an organoleptic perspective, the predominance of sesquiterpenes gives the oil a heavy, warm, and persistent fragrance, characterized by woody and balsamic notes, in contrast to the fresh and volatile aroma typical of monoterpene-rich oils. This quality makes the oil potentially valuable for use in perfumery formulations where "base notes" are desired (**Baser & Buchbauer, 2015**).

IV. Conclusion

This study presents, to the best of our knowledge, the first comprehensive characterization of the essential oil extracted from the leaves of *Piper guineense* from Côte d'Ivoire, using hydrodistillation, GC/MS, and ¹³C NMR analyses. The obtained yield (2.00%) and the identification of 41 compounds across five chemical classes demonstrate the remarkable metabolic diversity of this species. The overwhelming predominance of sesquiterpenes (92.46%) and the high combined proportion of hydrogenated azulene (20.54%) and β-caryophyllene (19.54%) constitute a distinctive chemical profile. The presence of these well-documented molecules, known for their anti-inflammatory, antioxidant, and immunomodulatory properties, provides a strong chemical basis for the traditional uses of *P. guineense*.

Nevertheless, to translate this potential into practical applications, further studies are required, including *in vitro* and *in vivo* pharmacological testing, isolation and comparative evaluation of major compounds, toxicity and mechanism-of-action studies, and the development of optimized formulations. Such research will not only provide scientific validation for the traditional medicinal uses of *P. guineense* but also promote the valorization of this local plant resource in high-value therapeutic and cosmetic products.

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