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Chemical composition of the essential oil extracted from the seeds of *Monodora myristica* (Gaertn.) dunal (Annonaceae) harvested in the mountain district in Côte d'Ivoire

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Abstract
As part of a contribution to the valuation of aromatic and medicinal plants of the Ivorian flora, our interest focused on the extraction and determination of the chemical composition by GC / MS of the essential oil has been extracted from the seeds of *Monodora myristica*. The yield of the essential oil is (1.43±0.08) %. Chemical analysis of the essential oil has identified 32 compounds. This corresponds to 99.96% of the total constituents. The phytochemical composition consists of hydrocarbon monoterpenes (77.47%), oxygenated monoterpenes (11.43%), hydrocarbon sesquiterpenes (6.54%), 0.55% oxygenated sesquiterpenes and other compounds (3 .99%). The chemical composition is dominated by hydrocarbon monoterpenes (77.47%). The major phytocompounds are ortho-cymene (39.54%), α-phellandrene (25.81%), α-pinene (5.6%) and α-terpineol (5.51%). The presence of phenolic hydroxy compounds, alcohol and sesquiterpenes would justify the use of the species in traditional medicine.

Keywords: *Monodora myristica*, essential oil, ortho-cymene, α-phellandrene and sesquiterpene

1. Introduction
Medicinal plants constitute a precious heritage for humanity and more particularly for the majority of poor communities in developing countries who depend on them for their primary health care and their livelihoods [1]. Their importance no longer needs to be demonstrated, especially in rural areas where, most often, they play a fundamental role in the survival and development of populations [2]. Despite their interesting properties, aromatic medicinal plants are nevertheless underestimated by populations. Therefore, more in-depth research on the essential oils extracted from these plants is necessary for a better knowledge of their chemical compositions and their physicochemical properties in order to derive the most benefits from them [3]. This knowledge linked to them constitutes a cultural value and can eventually allow the synthesis of new pharmaceutical drugs [4]. These multiple uses of plant species and essential oils have prompted researchers to study them with the aim of discovering new active molecules [5].

It is in this context that we became interested in *Monodora myristica*, an aromatic species and essential oils used by populations in traditional medicine. The species grows in the sub-Saharan African region. It is a tree with a fluted trunk at the base that can reach 25 m in height. Its fruits contain gray and fragrant seeds [6]. The almond of the grains of the species, rich in lipid, is edible. It is used as a spice because of its flavor and aroma in different culinary preparations [7, 8]. *M. myristica* (Gaertn.) Dunal, is a plant species widely used in traditional medicine. The juice from the pulped bark is used as a brush to treat scabies. It is sometimes prescribed in a steam bath to relieve fatigue and to treat feverish aches [6]. In Benin, the paste obtained after pounding the grains of *Monodora myristica* in association with those of *Xylopia aethiopica*, ginger rhizomes, red oil and diesel oil is used by massage in the commune of Adjarra in the south-east to treat the curvature [9]. The decoction of the seeds of the species is used by the populations to treat buruli ulcer. It is prescribed to wash the wound [10]. The decoction of the fruits is also indicated to treat infections, cysts when that of the roots can treat myomas and fibroids [4]. The crushed seeds of the species are used in the treatment of female sterility in the departments of Ouémé. The decoction is used orally to treat infertility and myomas. The macerated is prescribed for personal hygiene [11]. The extracts of the species are endowed with anti-inflammatory, antioxidant, anticancer and hypotensive activity [12]. The essential oils (EOs) have been the subject of several studies. In Côte d'Ivoire, the EO of the seeds harvested in Bondoukou extracted by hydrodistillation has a total chemical composition

of 91.02%. It is dominated by hydrocarbon monoterpenes 71.32%, followed by oxygenated monoterpenes (8.57%) and other compounds 10.36%. The hydrocarbon sesquiterpenes are in low proportions 0.77%. Oxygenated sesquiterpenes are absent. The main compounds are α-phellandrene (24.4%) and p-cymene (32%) [13]. The yield of the essential oil extracted by hydrodistillation from the dried seeds, harvested in September south of Bangui in Central Africa, is 1.2%. Its chemical composition is dominated by hydrocarbon monoterpenes (93.2%) including 77.4% hydrocarbons and 15.8% oxygenated. Sesquiterpenes are in low proportions 5.8% including 3% oxygenated and 2.8% hydrocarbons. The majority compounds are α-phellandrene (34.4%) and p-cymene (22.2%) The hypotensive activity of the essential oil of the species is of the same order as that of the reference drug used to lower blood pressure (verpamil) [14]. The EO, from the fruits of the species harvested in the south and west of Cameroon, extracted by hydrodistillation with a Clevenger-type device for 4 hours, has a yield of 2.33%.The chemical composition is dominated by hydrocarbon monoterpenes (91.4%). Oxygenated monoterpenes (5.1%), hydrocarbon sesquiterpenes (1%) and oxygenated sesquiterpenes (2%) are in low proportions. The majority compounds are α-phellandrene (57.8%) and p-cymene (12.7%) [15]. The EO of the grains of the Nigerian species extracted by hydrodistillation has a yield of 2.16%. The major compounds are germacrene D-4-ol (25.48% and linalool (15%) [16].

This is why, in order to contribute to a better valorization of the aromatic and medicinal plants of the Ivorian flora, we propose in this work, to extract and characterize by GC/MS the chemical composition of the essential oil of *Monodora myristica*.

2. Materials and Methods

2.1 Plant material

The plant material consists of the grains of *Monodora myristica* (Annonaceae). The fruits have been harvested in February 2022 in Kassiapleu, a village located in the department of Man more precisely in the Autonomous District of the mountains in Côte d'Ivoire. The plant has been identified thanks to the herbarium of the Center National de Floristique de Côte d'Ivoire (CNF) of the Félix Houphouët-Boigny University (Abidjan/Cocody) under the number H UCJ 001303.

2.2 Methods

2.2.1-Method of extraction of essential oils

The extraction has been carried out by hydrodistillation using a Clevenger-type device. To extract the essential oil from the seeds of the species, we used a 6-liter round bottom flask containing approximately 3 L of distilled water, plant material and a condenser. The grains have been pounded. A cooler is mounted on the flask containing the plant material. The whole thing is heated using a hot plate. The volatile compounds are carried with the water vapor into the condensation column. The floral water and the essential oil constitute the vapor condensed in a binary azeotropic mixture. The two constituents are separated by decantation under the effect of their densities. The essential oil is dried over anhydrous sodium sulfate. All collected essential oil samples are placed in tinted bottles protected from light using aluminum foil and then stored in the refrigerator at a temperature of -9°C [17, 18].

2.2.2 Determination of chemical composition

A gas chromatograph (7890A, Agilent Technologies) combined to a mass spectrometer (5975C, Agilent Technologies) was used to analyze EO. In fact, the EO was diluted in dichloromethane (1:100). One microliter of EO was injected into an HP-5MS capillary column at a temperature of 250°C. The oven temperature increased to 40°C for 5 min, then at 2°C/min for 15 min up to 250°C, with a flow rate of 10°C/min up to 300°C. The carrier gas was helium with a flow rate of 1 mL/min. At a temperature of 280°C, the MS detector had a voltage of 1.4 kV. Ions with m/z ratios greater than 40 and less than 500 could be detected. The data collected after GC MS analysis were compared with those in the literature and those in the NSIT (National Institute of Standards and Technology) database for the identification of compounds. [19, 20].

$$Ri = 100\left[n + \frac{t_R(C_i) - t_r(C_n)}{t_r(C_{n+1}) - t_R(C_n)}\right]$$

Ri : Retention index

Ci: Unknown compound of HE;
Cn: linear alkane (comprising n C atoms) whose retention time precedes the unknown EO compound;
Cn+1: Linear alkane (containing n C atoms) the unknown compound;
n: carbon number of the linear alkane.
tR(Cn) retention time of the linear alkane with n carbon atoms.

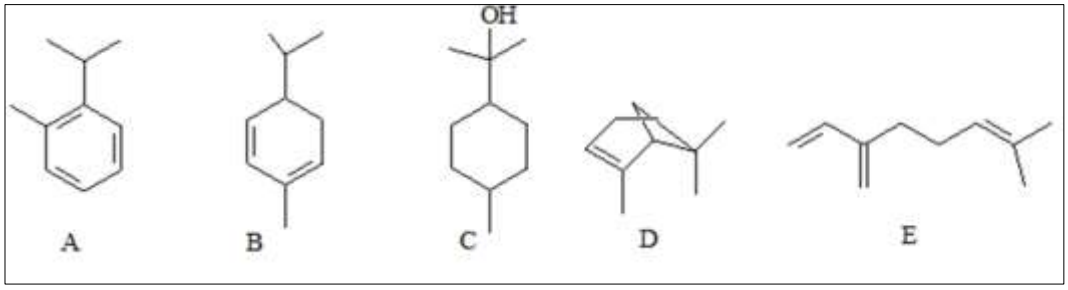
3. Results and Discussion

3.1 Result of the extraction

The essential oil obtained by hydrodistillation has a pale yellow color with an aromatic smell. The yield of essential oil extracted from the seeds of *M. myristica* is (1.43±0.08)%. Our results are different from those reported in the literature [14-16]. These found returns ranging from 1.2 to 2.33%. The observed differences in essential oil yield from one organ to another or from one species to another have been reported in the literature. According to several authors, the origin of the harvest of the species, the harvest period, the organ of the plant, the duration of drying and the method of extraction are factors among many others that can also have a influence on essential oil yields [21-27].

3.2. Phytochemical composition of essential oils extracted by GC-MS from organs

Analysis of the chromatogram and mass spectra of the essential oil from *M. myristica* identified 32 compounds. This corresponds to 99.96% of the total constituents of the essential oil. These results are recorded in Table 1. It consists of hydrocarbon monoterpenes (77.47%), oxygenated monoterpenes (11.43%), hydrocarbon sesquiterpenes (6.54%), 0.55% oxygenated sesquiterpenes and other compounds (3.99%). Hydrocarbon monoterpenes are widely present with 77.47%. Ortho-cymene (A; 39.54%), α-phellandrene (B; 25.81%), α-pinene (C; 5.6%), α-terpineol (D; 5.51%) and β-myrcene (E; 3.21%) are the major phytocompounds. The structures of the majority constituents are represented in figure 1 and their proportions in figure 2.



A: Ortho-cymene; B: α-phellandrene; C: α-pinene; D: α-terpineol and E: β-myrcene

Fig 1: Structure of the main compound of the EO of *M. myristica*

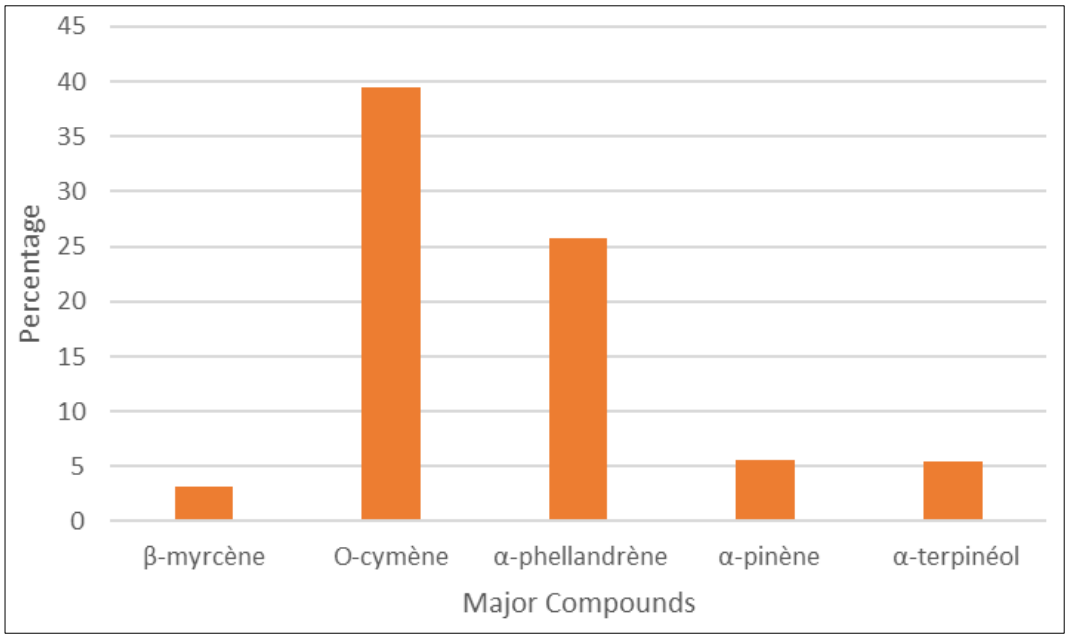


Fig 2: Parentage of majority compound

Previous studies regarding the phytochemical composition of the volatile oils of *M. myristica* have been carried out. The results of these authors established that the essential oils of the seeds of the species are also dominated by hydrocarbon monoterpenes with the respective proportions of 71.32%, 77.4% and 91.4% [13-15].

By comparing our result to previous work, we note a similarity in chemical composition but with different proportions. This similarity of results could be explained by the fact that these countries are located in the tropical zone. The majority phytoconstituent of the studies reported in the literature is α-phellandrene in variable contents respectively 24.4%, 34.4% and 57.8%. It is different from that of our study (ortho-cymene) according to several authors [14,

Table 1: Chemical composition of the essential oil of *Monodora Myristica* seeds

N°	Rt	Ri	Compounds	M/Z	Content
1	12.27	919	α-thujene	136	1.96
2	12.58	924	α-pinene	136	5.6
3	15.31	966	β-pinene	136	0.26
4	16.69	988	β-myrcene	136	3.21
5	17.33	998	α-phellandrene	136	25.81
6	18.20	1010	(+)-4-carene	136	0.19
7	18.80	1818	Ortho-cymene	136	39.54
8	20.05	1036	(E)-β-ocimene	136	0.34
9	21.27	1053	γ-terpinene	136	0.40
10	24.41	1096	Linalol	154	2.65
11	26.66	1115	ci-P-menth-2-en-1-ol	154	0.36
12	26.97	1133	3-carene	136	0.16
13	30.57	1184	endo-borneol	154	0.72
14	31.29	1194	α-terpineol	154	5.51
15	31.76	1201	Safranal	150	0.37
16	32.74	1216	Isomaltol	126	0.2
17	33.82	1232	Formate de bornyle	182	0.43
18	37.96	1293	Thymol	150	0.65
19	38.53	1301	Carvacrol	150	1.17

20	45.37	1408	β-Caryophyllene	204	0.60
21	47.45	1442	α-Caryophyllene	204	0,14
22	48.26	1455	4'-pentylacetophenone	190	2.97
23	48.97	1467	PrecoceneI	190	0.29
24	49.43	1475	Cadina-1(6),4-diene trans	204	0.27
25	50.05	1485	α-elemene	204	0.33
26	50.46	1492	Bicyclogermacrene	204	0.29
27	51.23	1505	α-muurolene	204	0.72
28	51.83	1515	γ-cadinene	204	1.75
29	54.74	1565	Epizonarene	204	1.95
30	58.52	1633	Tau-cadinol	222	0.53
31	59.23	1646	Cadin-1(10),4-diene	204	0.49
32	106.67	2735	Bis(2-éthylhexyl)phtalate	390	0.10
			Hydrocarbon monoterpenes		77.47
			Oxygenated monoterpenes		11.43
			Hydrocarbon sesquiterpenes		6.54
			Oxygenated Sesquiterpenes		0.55
			Other		3.99
			Total		99.96

Rt: retention time; Ri: retention index; M/Z: specific load

Conclusion

As part of the development of aromatic and medicinal plants of the Ivorian flora, this study made it possible to determine the yield and the chemical composition of *Monodora myristica*. The yield of essential oil extracted from the seeds (1.43±0.08) % is acceptable. The chemical analyzes by GC/MS made it possible to identify 32 phytocompounds in the essential oil of the species. It is dominated by hydrocarbon monoterpenes (77.47%). The majority phytocompounds are ortho-cymene (39.54%) and α-phellandrene (25.81%), Thymol and carvacrol are powerful bactericides and anti-infectives. They also help relieve muscle pain and fight general fatigue. Their presence in the essential oil of *M. myristica* would justify the use of the species in traditional medicine.

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