

Leaf Epicuticular Wax Chemical Composition of *Morettia phillaeana* (Del.) DC.**From Sudan**

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Abstract

Morettia phillaeana was analyzed for its chemical composition of surface epicuticular wax. The constituents were extracted from the leaves by using n-hexane solvent, and were identified by using Gas chromatography-mass spectroscopy (GC/MS). The major components were hydrocarbons (74.15%), fatty acids, methyl ester (15.73%). and phytosterol (5.47%). Twelve components out of 29 were characterized representing 90.98% of total components. The most abundant compounds were tetracontane (57.74%), Octacosane (6.58%), β -sitosterol (5.47%), Hexadecanoic acid, methyl ester (3.81%), pentacosane (3.77%) and 2-methylhexacosane (3.43%).

Keywords: Chemical composition, Epicuticular Wax, *Morettia phillaeana*, Sudan

Introduction;

Epicuticular surface wax of higher plant and their organs are very often covered by thin layer of wax that are in fact mixtures of long chain of fatty acids, esters, alcohols, hydrocarbons and others classes of compounds [1]. The composition of epicuticular wax could be a potential marker for species groups reflecting both ecological and genetic relationships [2]. Surface wax are first plants barrier encountered by any material applied to foliage. For a compound to penetrate into the plant it must spread over the surface of the leaf, stem, or fruit, and then pass through the surface wax [3]. The wax on the external surface of plants (cuticular or epicuticular wax) is a complex mixture of aliphatic lipid compounds, like sugar and amino acids as well as secondary metabolites and the

chemical composition of which differs greatly among different plant species and, to a lesser extent, between different parts of the plant. Leaves and floral parts of the plant tend to have the highest concentrations of wax. The epicuticular wax is the contact point between plant and environment and plays crucial role in mediating biotic and abiotic interaction [4].

Morettia phillaeana (Del) DC. (Brassicaceae) is one of two *Morettia* species which occur in Sudan [5]. It grow in the northern and central region of the country, where it is known by the popular name of “Gabshah. Its smell is characteristic. Stiff erect hispid herb. Leaves linear to lanceolate, hoary or hispid. Petals pinkish-white, slightly longer than the very hairy sepals. Fruit strongly curved [6] - [7].

The present investigation was carried out to determine the possible chemical components from n-hexane extract of *M.phillaeana* of leaf epicuticular wax using Gas Chromatography/Mass Spectrometry.

Materials and Methods :

Plant material:

Leaves of *Morettia phillaeana* were collected from a flowering plant during winter time in 2017 from University Campus, Omdurman Islamic University in the Omdurman south, Khartoum State Central Sudan. Hatil H. EL-Kamali was carried out taxonomic authentication of the plant.

The Wax extraction for surface n-hexane soluble compounds:

Epicuticular surface wax was extracted by immersing the plant sample (leaves) in n-hexane for 90 secs. This procedure , extracts only surface n-hexane-soluble compounds. n-hexane extract was filtered and concentrated by evaporation. The constituents of the epicuticular wax were characterized and identified by GC/MS [8].

Gas Chromatography-Mass Spectrometry Analysis of the compounds:

The constituents of the Epicuticular wax were characterized and identified by GC/MS. GC/MS analysis were performed on_a shimadzu GC/MS-QP2010A system in ET mode (70ev) equipped with a split /splitters injector (250°C) , at split ratio of 5/50 using DB-5MSColumn (30m x 0.25mm , film thickness: 0002E25 miss J and W scientific , fulsome , CA,WA). Injection volume was 1misar litre and electronic pressure programming was used to maintain a constant flame (0.67 mL/min) of the Helium carrier gas. The even temperature was programmed from 150°C (4mins) to 320°C at a rate of 2 cm³/min and held at temperature 200°C and interface temperature 250°C. The relative approach percentage of each compound was determined by area under the peak. Components identification was carried out using the NIST 147 and NIST 27 libraries.

Results and Discussion:

Surface Epicuticular wax from leaves of *Morettia phillaeana* was extracted using n-hexane as a solvents for 90 secs. This procedure extracted only surface n- hexane soluble compounds. The compound reported from *Morettia phillaeana* n-hexane extract have not been reported in literature before .The GC/MS chromatogram (Figure 1) of the n-hexane extract revealed the presence of 29 compound six compounds were characterized representing 80.80% of the total components (Figure 2) The identified compounds, Area%, Formula, Class type, are shown in Tables 1 and 2. The most abundant compounds were Tetracontane (57.74%),Octacosane (6.58%) , β _sitosterol (5.47%) , Hexadecanoicacid,methylester (3.81%) , Pentacosane (3.77%) , and 2-methylhexacosane(3.43%).

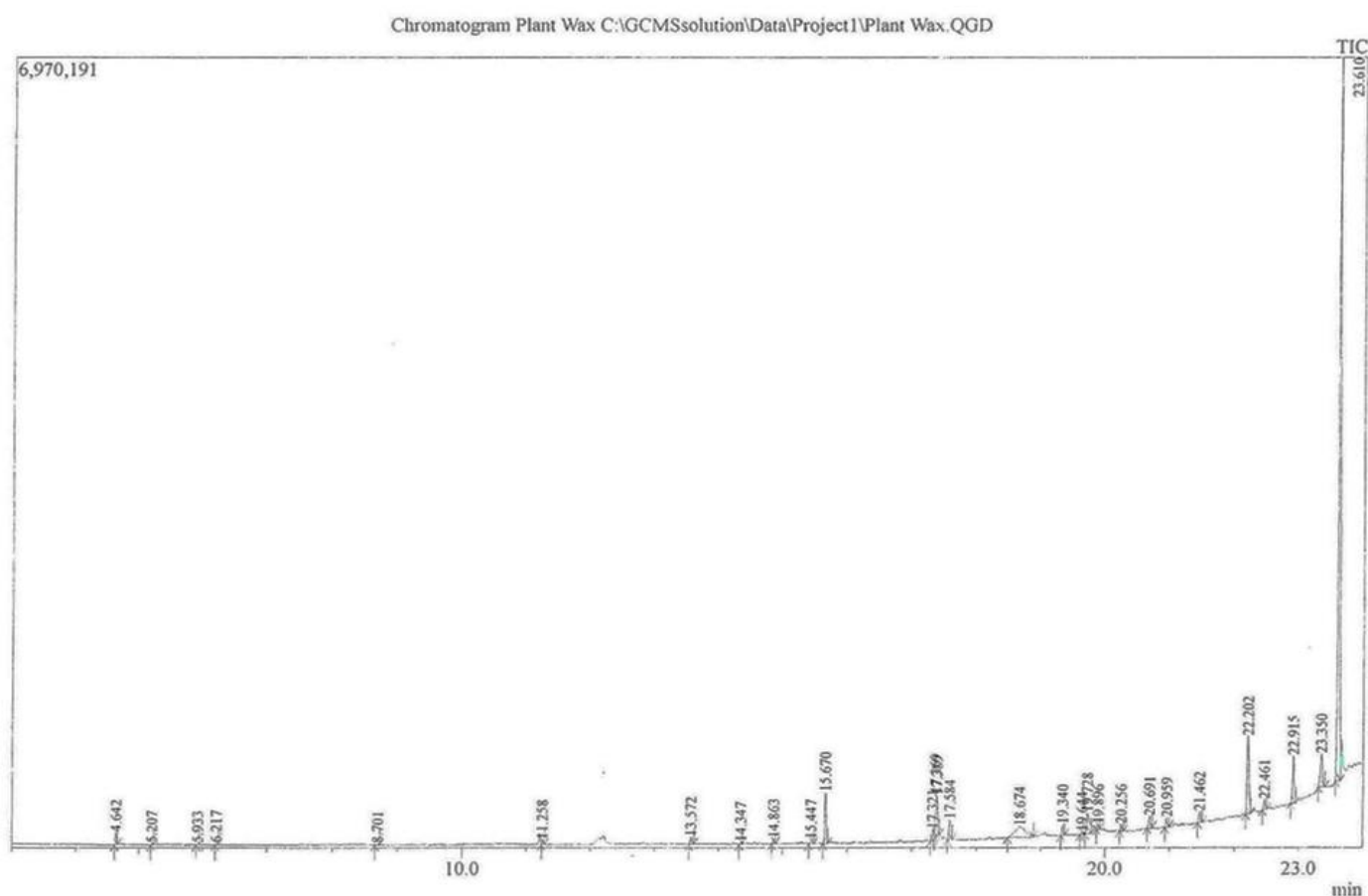


Figure1. The GC/MS chromatogram of *M. phillaeana* epicuticular wax n-hexane extract

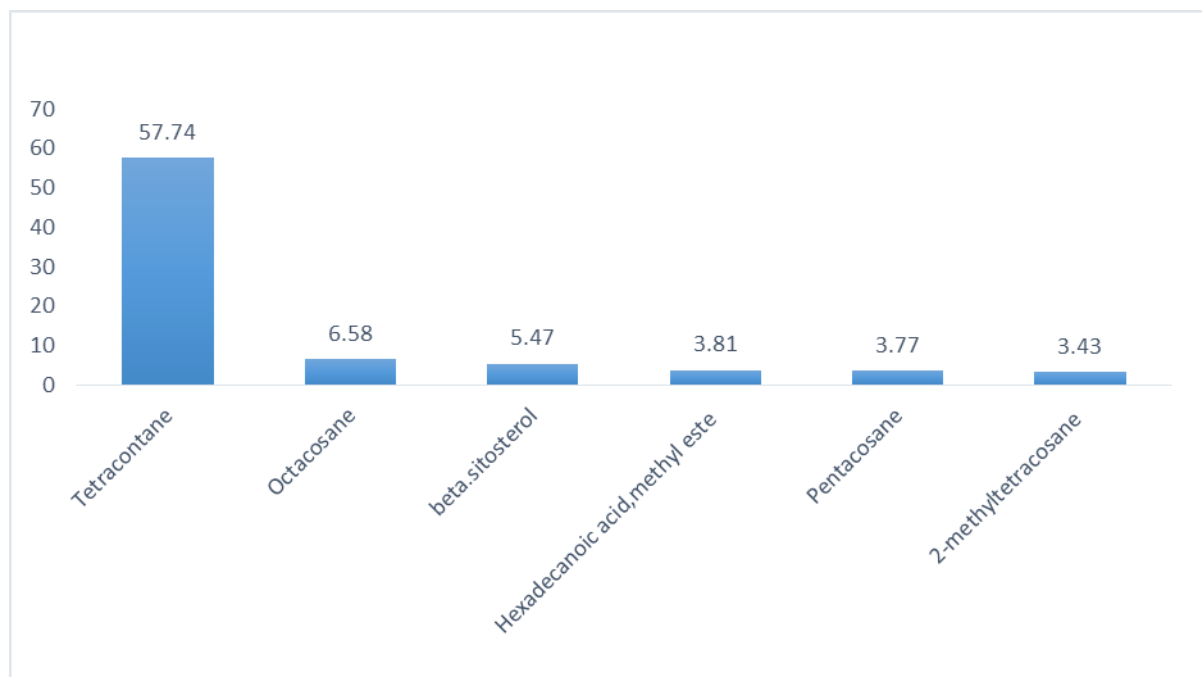


Figure 2. Major components from n-hexane extract of epicuticular leaves wax of *M. phillaeana*

The constituents of the n-hexane extract of surface epicuticular wax from leaves of *Morettia phillaeana* belong to seven classes of compound including hydrocarbons (74.15%) , fatty acids methyl ester (15.73%) , alkenamide (2.83%) , phytosterols (5.47%) , alcohols (0.98%) , ketones (0.34%) and aromatic hydrocarbons compounds (0.49%).

Table 1. Composition of surface epicuticular wax of *Morettia phillaeana*

No	Compound	Formula	Area%
1	1-hexanol,2-ethyl	C ₁₈ H ₁₈ O	0.74
2	1-octanol	C ₈ H ₁₈ O	0.12
3	Octanoic acid, methyl ester	C ₉ H ₁₈ O ₂	0.08
4	(S)-(+)-6-methyl-1-octanol	C ₉ H ₂₀ O	0.12
5	Decanoic acid methyl ester	C ₁₁ H ₂₂ O ₂	0.11
6	Dodecanoic acid, methyl ester	C ₁₃ H ₂₆ O ₂	0.29
7	Methyl tetradecanoate	C ₁₅ H ₃₀ O ₂	0.67
8	Octadecane	C ₁₈ H ₃₈	0.13
9	2-pentadecanone, 6,10,14-trimethyl	C ₁₈ H ₃₆ O	0.34
10	8,11,14-Docosatrienoic acid,methyl ester	C ₂₃ H ₄₀ O ₂	0.42
11	Hexadecanoicacid,mehtyl ester	C ₁₇ H ₃₄ O ₂	3.81
12	9,12-octadecadienoic acid (Z,Z)-,methyl	C ₁₉ H ₃₄ O ₂	0.94
13	9-octadecanoic acid,mehtylester,(E)-	C ₁₉ H ₃₆ O ₂	2.89
14	9,12,15-octadecatrienoic acid, methyl ester	C ₁₉ H ₃₂ O ₂	2.42
15	Methyl stearate	C ₁₉ H ₃₈ O ₂	1.49
16	Beta.sitosterol	C ₂₉ H ₅₀ O	5.47
17	Eicosanoic acid, methyl ester	C ₂₁ H ₄₂ O ₂	0.82
18	4,8,12,16-tetramethyl heptadecan-4-olide	C ₂₁ H ₄₀ O ₂	0.20
19	9-octadecenamide,(Z)-	C ₁₈ H ₃₅ NO	2.83
20	Octacosane	C ₂₈ H ₅₈	0.39
21	Phenol,2,2-methylenebis[6-(1,1-dimethyl)-4-mehtyl]	C ₂₃ H ₃₂ O ₂	0.49
22	Pentatricontane	C ₃₅ H ₇₂	1.13
23	Docosanoic acid, methyl ester	C ₂₃ H ₄₆ O ₂	0.73
24	Tetratetracontane	C ₄₄ H ₉₀	0.98
25	Octacosane	C ₂₈ H ₅₈	6.58
26	Tetracosanoic acid, methyl ester	C ₂₅ H ₅₀ O ₂	0.86
27	Pentacosane	C ₂₅ H ₅₂	3.77
28	2-methylhexacosane	C ₂₅ H ₅₂	3.43
29	Tetracontane	C ₄₀ H ₈₂	57.74

Table 2. Constituents of epicuticular wax of *Morettia phillaeana* according to chemical class

Hydrocarbons		
Alkanes		
No	Compound	Area%
1	Octadecane	0.13
2	Octacosane	0.39
3	Pentatricontane	1.13
4	Tetratetracontane	0.98
5	Octacosane	6.58
6	Pentacosane	3.77
7	2-methylhexacosane	3.43
8	Tetracontane	57.74
		74.15
Fatty acids		
1	Octanoic acid, methyl ester	0.08
2	Decanoic acid methyl ester	0.11
3	Dodecanoic acid, methyl ester	0.29
4	Methyl tetradecanoate	0.67
5	8,11,14-Docosatrienoic acid,methyl ester	0.42
6	Hexadecanoicacid,mehtyl ester	3.81
7	9,12-octadecadienoic acid (Z,Z)-,methyl	0.94
8	9-octadecanoic acid,mehtylester,(E)-	2.89
9	9,12,15-octadecatrienoic acid, methyl ester	2.42

10	Methyl stearate	1.49
11	Eicosanoic acid, methyl ester	0.82
12	4,8,12,16-tetramethyl heptadecan-4-olide	0.20
13	Docosanoic acid, methyl ester	0.73
14	Tetracosanoic acid, methyl ester	0.86
		15.73
Alcohols		
1	1-hexanol,2-ethyl	0.74
2	1-octanol	0.12
3	(S)-(+)-6-methyl-1-octanol	0.12
		0.98
Ketone		
1	2-pentadecanone, 6,10,14-trimethyl	0.34
Amide		
1	9-octadecenamide,(Z)-	2.83
Aromatic hydrocarbons		
1	Phenol,2,2-methylenebis[6-(1,1-dimethyl)-4-mehtyl]	0.49
Phytosterol		
1	Beta.sitosterol	5.47

Higher percentage of hydrocarbons was found in the *Morettia* extract , which might be used in the fuel industry. The biological activities of some of the components beta-sitosterol may have the potential for prevention and therapy for human cancer [9]. Fatty acids can attribute to

antioxidant activity , palmitic acid was reported to be a more effective free radical scavenger than beta-carotene [10] and octacosane it have mosicidal activity [11] while tetracontane may have the potential as hypoglycemic and antioxidant [10] , and these can be used in pharmaceutical formulations.

The usefulness of leaf wax alkanes as chemotaxonomic characters at the familial , sub-familial and tribal levels in plant families Apiaceae, Brassicaceae and Leguminosae. Alkanes from C16) upwards form the most important components of fuel oil and lubricating oil, as anti-corrosive agents and as their hydrophobic nature means that water cannot reach the metal surface. *M.phillyaeana* was characterized by the presence of n-alkane composition C44, C40, C35, C28 and C25. Long chain n-alkanes (C21-C37) are among the most long-lived and widely utilized in terrestrial plant biomarkers [12].

Conclusion :

The components reported from *M.phillyaeana* n-hexane extract have not been reported in literature before. The GC/MS chromatogram of the n-hexane extract revealed the presence of 29 compounds. Twelve components were characterized representing 90.98% of the total components.

Recommendations:

We hope to confirm of those identified bioactive compounds by using better and more sophisticated devices than GC/MS.

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