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Wax Lipids from Leaf Surfaces of Some Common plants of Malaysia

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ABSTRAK

Ekstrak lipid lilin epikutikular daripada daun-daun beberapa jenis tumbuhan yang terdapat di Lembah Klang telah di asingkan kepada pecahan-pecahan hidrokarbon, asid karboksilik, keton dan alkohol, dan seterusnya dianalisis dengan menggunakan kaedah kromatografi gas (KG) dan kromatografi gas spektrometri jisim (KG-SJ). *Mesau ferrea* menghasilkan amaun lilin terbanyak, sementara yang paling sedikit diperolehi daripada *Eugenia grandis.* Dalam kebanyakan spesies yang dikaji, *n*-alkana utama yang ditemui ialah hentriakontana (C_{31}), diikuti oleh triakontana (C_{33}) dan nonakosana (C_{29}). *n*-Alkana mempamerkan pola mata-gergaji, iaitu merupakan satu ciri hidrokarbon dari sumber biogenik, sementara asid *n*-alkanoik dan *n*-alkanol pula mempamerkan satu corak di mana bilangan karbon genap lebih ketara daripada bilangan karbon ganjil. β-Sitosterol merupakan fitosterol utama yang dikesan di dalam sesetengah spesies, tetapi dalam kuantiti surihan sahaja. Triterpenoid dan triterpena kebanyakannya ditemui di dalam lilin spesies *Mesua ferrea* di mana asid α- dan β-boswelik merupakan komponen utama, dan α- dan β-amyrin dengan amaun yang minor. Biopenunjuk lain yang dikesan termasuklah α- dan β-amyron, friedelin, friedelanol, friedelana, olean-12-ena, tarakserena, skualena, asid dihidroniktantik, asid dihidroburik dan asid dihidrokanarik. Kajian ini menunjukkan bahawa taburan n-alkana merupakan fungsi suhu persekitaran setempat.

ABSTRACT

Epicuticular wax lipids from leaves of some common plant species found in the Klang Valley, Malaysia were extracted into dichloromethane for analysis. Extracts were separated into hydrocarbon, carboxylic acid, ketone and alcohol fractions. The fractions after derivatization were then subjected to gas chromatographic (GC) and gas chromatography-mass spectrometric (GC-MS) analyses. The wax yield was highest for *Mesua ferrea* while the lowest yield was obtained from *Eugenia grandis.* In most species studied the major *n*-alkane was found to be hentriacontane (C_{31}), followed by tritriacontane (C_{33}) and nonacosane (C_{29}), respectively. The *n*-alkanes exhibit a saw-tooth pattern which is characteristic of a biogenic origin. A strong even-to-odd carbon number predominance is observed for both the *n*-alkanoic acids and *n*-alkanols. Only traces of mainly β-sitosterol were detected in some of the species. The triterpenoids and triterpenes were mostly found in the wax of *Mesua ferrea* with α- and β-boswellic acids as the major components and minor amounts of α- and β-amyrin. Other biomarkers identified were α- and β-amyrones, friedelin, friedelanol, friedelane, olean-12-ene, taraxerene, squalene, dihydronyctanthic acid, dihydroroburic acid and dihydrocanaric acid. This study has shown that the distribution of *n*-alkanes is a function of the ambient temperature.

KeyWords: epicuticular wax, organic chemical analysis, GC, GC-MS

INTRODUCTION

The leaf surfaces of vascular plants generally consist of a lipophilic polymer membrane (cuticle) whose monomers are mostly hydroxy fatty acids (Kolattukudy, 1970, 1980; Martin and Juniper, 1970). Growing from the cuticular membrane are complex mixtures of lipids, often in crystalline form, called epicuticular plant waxes. These waxes act as a protective barrier between cuticula and the atmosphere (Eglinton *et al.* 1962a; Hall and Jones, 1961; Jameison and Reid, 1972; Kolattukudy, 1970, 1976; Martin and Juniper, 1970; Rogge *et al.* 1993a; Schreiber and Schönherr, 1992) and also to prevent losts of water, ions, etc. from the leaves.

Epicuticular plant waxes consist mainly of aliphatic compounds such as higher molecular weight (usually $>C_{24}$) *n*-alkanes, *n*-alkanals, *n*-alkanols, *n*-alkanoic acids, and wax esters (Eglinton *et al.* 1962a; Kolattukudy, 1970, 1976; Simoneit and Mazurek, 1982; Simoneit, 1989). Due to wind-induced mechanical shear, rubbing of leaves against each other and rainfall, such epicuticular wax protrusions become airborne. In some areas, the leaf material produced during an annual growing cycle is deposited in nearby gardens and along streets. In this manner, leaf waxes may be added to garden soil and to road dust. Wind- or traffic-induced turbulence can resuspend garden soil and road dust, thus entraining particles of vegetative detritus into the atmosphere. Therefore, it is not uncommon to find the occurrence of plant wax constituents in airborne particulate matter in both urban and rural aerosols (Doskey and Andren, 1986; Gagosian *et al.* 1982; Mazurek *et al.* 1991; Rogge *et al.* 1993b; Sicre *et al.* 1990; Simoneit, 1979, 1989; Simoneit and Mazurek, 1982; Simoneit *et al.* 1988, 1990, 1991b; Wils *et al.* 1982). So any study on the organic composition of atmospheric aerosols will require knowledge of the natural background signal from indigenous vegetation.

Many compositions of plant waxes from individual species have been reported in order to utilize the data for chemotaxanomic correlations (e.g. Eglinton *et al.* 1962a,b; Eglinton and Hamilton, 1963; Borges del Castillo *et al.* 1967; Herbin and Robins, 1968a,b; Martin-Smith *et al.* 1967; Nishimoto, 1974; Salasoo, 1987; Stránsko(y,´) *et al.* 1967; Tulloch, 1976; Wollrab, 1967; Zygaldo *et al.* 1994). However, the plant wax composition found in aerosols is an admixture, generally representative of the plant species in the air shed

(Simoneit, 1977, 1989; Simoneit and Mazurek, 1982). Thus it is more useful to consider vegetation species which are representative of a particular study area for wax analysis, in order to use the results for aerosol source reconciliations.

This paper describes the composition of the solvent extractable organic matter obtained from leaf surfaces of ten plant species commonly found in and around the city of Kuala Lumpur, Malaysia. It is hoped that this study will enable us to identify some suitable marker compounds or marker compound assemblages for tracing vegetative detritus particularly in the atmosphere of Kuala Lumpur.

EXPERIMENTAL

Ten plant species were chosen for this study based on their high abundance in and around the city of Kuala Lumpur. The plants were sampled mostly on the campus of the Universiti Malaya. The species chosen were: *Calophyllum inophyllum* Linn. (Guttiferae); *Cerbera odollam* Gaertn. (Apocynaceae); *Eugenia grandis* Wight (Myrtaceae); *Fagraea fragrans* Roxb. (Loganiaceae); *Hevea brasiliensis* M.A. (Euphorbiaceae); *Melaleuca leucadendron* Linn. (Myrtaceae); *Mimusops elengi* Linn. (Sapotaceae); *Mesua ferrea* Linn. (Guttiferae); *Lagerstroemia indica* Linn. (Lythraceae); and *Pterocarpus indicus* Willd. (Leguminoseae). Descriptions of these species can be found in Burkill (1935).

Sample Extraction

A representative sample of between 20-45 mature leaves of uniform size and appearance was taken from at least five individual plants of a particular species and combined. Care was taken to exclude any insects from the leaves. The leaves were placed in a paper bag before being transported to the laboratory. In the laboratory, each leaf was dipped (up to the stem) for 3-5 seconds into about 150 ml of distilled-in-glass methylene chloride. All leaves were handled with forceps. The extracted leaves were dried and weighed. The excess solvent was removed by rotary evaporation and nitrogen blowdown to 1-2 ml. Each concentrated extract was transferred to a vial with Teflon lined cap and kept refrigerated until further analysis.

Lipid Separation and Analysis

The concentrated extracts were reacted with BF_3-methanol to esterify carboxylic acids and separated into hydrocarbon, ester, ketone (and aldehyde) and alcohol fractions by thin layer chromatography on silica gel plates. These fractions were then subjected to gas chromatographic (GC) and gas chromatography-mass spectrometric (GC-MS) analyses. Alcohol fractions were converted to the trimethylsilyl ethers prior to GC and GC-MS analysis by reaction with *N,O*-bis-(timethylsilyl)-trifluoroacetamide(BSTFA). Perdeutero-*n*-dotriacontane was used as a coinjection standard. A more detailed description of the complete experimental format has been described elsewhere (Abas *et al.* 1995). The GC analyses used a capillary column coated with DB-1 (30 m 3 0.25 mm i.d.) and the operating conditions, for the analyses of fraction 1 only, were as follows: isothermal at 65°C for 2 min, temperature program 65-180°C at 25°C per minute and 180-310°C at 4°C per minute, then hold isothermal at 310°C for 60 min. A slightly slower temperature programme (65 to 310°C at 4°C/min) was used for the other fractions.

The GC-MS analyses were conducted on a Finnigan Model 4021 quadrupole mass spectrometer interfaced directly with a Finnigan Model 9610 gas chromatograph equipped with the same column as for the GC analyses. The GC used the same operating conditions as above and the MS scanned for 50-650 da at 2 sec/decade with an electron impact ionization potential of 70 eV. Mass spectrometric data were acquired and processed with the Finnigan-Incos data system. In GC-MS analyses the homologous compound series are defined by key ion mass chromatograms. Thus, the *n*-alkanes are characterized in m/z 99 fragmentograms, the *n*-alkanoic acids as the methyl esters in m/z 74 or 87 fragmentograms, the *n*-alkanols as the trimethylsilyl ethers in m/z 75 fragmentograms and by their M-15 ions, and the *n*-alkanals and *n*-alkan-2-ones in m/z 71 and m/z 58 fragmentograms, respectively. Alkyl diesters of benzene-1,2-dicarboxylic acid (phthalates) are identified by m/z 149 and m/z 163 fragmentograms.

RESULTS AND DISCUSSION

The extract yields and lipid characteristics of the plant waxes from the species studied are given in Table 1. The wax yield was highest for *Mesua ferrea* while the lowest yield was obtained from *Eugenia grandis.* Because of the similarity of the compound series amongst the species, only a selected number of samples was examined further by GC-MS. The carbon number maxima (C_{max}), carbon number range and carbon preference indices (CPI) for the *n*-alkanes, *n*-alkanoic acids and *n*-alkanols are also listed in Table 1.

Aliphatic Hydrocarbons

The *n*-alkanes were identified in fraction 1 of the separated wax extracts and are listed in Table 2. The *n*-alkanes of biosynthesized leaf wax exhibit a strong odd-to-even carbon number predominance with the dominant *n*-alkanes being C_{29}, C_{31}, and C_{33} (Eglinton *et al.* 1962b; Kolattukudy, 1970; Rieley *et al.* 1991; Simoneit and Mazurek, 1982). This is also evident in our study where we identified *n*-alkanes ranging from C_{19} to C_{36}, having the odd-carbon *n*-alkanes exceeding their even-carbon neighbors, thus exhibiting a saw-tooth pattern which is characteristic of biogenic *n*-alkanes. In most species the major alkane (C_{max}, Mazurek and Simoneit, 1984) was hentriacontane (C_{31}), followed by tritriacontane (C_{33}) and nonacosane (C_{29}), respectively. It was also noted that triacontane (C_{30}) was present as an important component in *Fagraea fragrans.* The C_{max} for the *n*-alkanes of vascular plant wax samples from Amazonia,

TABLE 1

Extract yields and lipid characteristics of epicuticular plant waxes investigated

			n-Alkanes				n-Alkanoc acids			n-Alkanols		
Plant Species1	Code	Extract yield (mg/g dry wt.)	yield (mg/g)	Fraction of total wax(%)	C_{max}[2]	CP[3]	C_{max}[2]	Range	CPI[3]	C_{max}[2]	Range	CPI[3]
Calophyllum inophyllum (All Heal Tree, Penaga Laut)	Cn	6.4	2.0	32	$\underline{29}$,31	13.3	$\underline{16}$,24,26	16-23	13.5	10,$\underline{16}$	10-20	∞
Cerbera odollum (Yellow-eyed Carbera, Pong-Pong)	Co	18.2	4.6	25	$\underline{31}$,33,35	13.5	16,$\underline{32}$,34	14-36	35.5	$\underline{34}$	24-36	18.3
Eugenia grandis (Sea Apple, Jambu Laut)	Eg	1.2	0.4	33	$\underline{31}$	14.5	16,$\underline{32}$,34	12-34	5.9	$\underline{32}$	24-34	12.2
Fagraea fragrans (Common Tembusu, Tembusu)	Ff	7.2	0.6	8	29,30,$\underline{31}$	3.6	$\underline{24}$,26,28	16-34	12.2	10,$\underline{28}$	10-34	6.2
Hevea brasiliences (Para Rubber Tree, Getah Para)	Hb	4.3	0.7	15	$\underline{31}$,33	7.2	$\underline{20}$,24	14-24	40.7	$\underline{18}$	18-20	∞
Lagerstroemia indica (Crepe Myrtle, Bungor)	Li	2.8	0.3	11	$\underline{31}$	6.5	$\underline{16}$,20,26	12-36	14.8	26,$\underline{28}$	22-36	8.5
Mimusops elengi (Tanjung Tree, Pokok Tanjung)	Me	2.0	1.6	79	31,$\underline{33}$	18.1	16,$\underline{28}$,30	14-34	26.6	$\underline{28}$,30	18-36	14.4
Mesua ferrea (Indian Rose Chestnut, Penaga Lilin)	Mf	35.4	0.04	2	29,$\underline{31}$	9.6	$\underline{16}$,18	14-32	37.2	$\underline{30}$,32	24-34	5.3
Melaleuca leucadendron (Capeput, Gelam)	Ml	5.6	0.04	0.6	$\underline{31}$	3.0	22,$\underline{28}$,30	14-32	7.3	28,$\underline{30}$	24-34	5.3
Pterocarpusindicus (Red Sandalwood. Angsana)	Pi	5.0	1.7	33	29,$\underline{31}$,33	25.2	$\underline{16}$,26	14-34	97.5	$\underline{26}$,28	22-36	30.1

[1] Latin name, in parentheses English common name, Malaysian vernacular name.

[2] C_{max} = carbon number maximum as defined in Mazurek and Simoneit (1984).

[3] CPI = Carbon preference index is the summation of the odd carbon number homologs divided by the summation of the even carbon number homologs for *n*-alkanes and the inverse for *n*-alkanoic acids and *n*-alkanols (Mazurek and Simoneit, 1984).

TABLE 2
Major *n*-alkanes and biomarkers in the epicuticular plant waxes analyzed from Malaysia

Compound		Plant Species									
Name	Composition	Cn	Co	Eg	Ff	Hb	Li	Me	Mf	Ml	Pi
n-Alkanes (relative concentration*)											
Docosane	$C_{22}H_{46}$	0.6			0.4						
Tricosane	$C_{23}H_{48}$	0.7			1.2	1			1	2	0.4
Tetracosane	$C_{24}H_{50}$	0.5			1	2	1	0.2	2	8	0.4
Pentacosane	$C_{25}H_{52}$	0.9			2	2	3	0.4	2	12	1.5
Hexacosane	$C_{26}H_{54}$	0.8			1	2	2	0.4	2	15	0.5
Heptacosane	$C_{27}H_{56}$	3		0.5	14	3	6	2.6	3	22	4.2
Octacosane	$C_{28}H_{58}$	3		0.5	7	2	3	0.5	2	19	0.7
Nonacosane	$C_{29}H_{60}$	100	5	16	78	9	36	15	32	41	33
Tiacontane	$C_{30}H_{62}$	7	1	3	37	4	10	2	6	11	2
Hentriacontane	$C_{31}H_{64}$	90	61	100	100	100	100	81	100	100	100
Dotriacontane	$C_{32}H_{66}$	3	10	5	9	16	13	7	3	9	3
Tritriacontane	$C_{33}H_{68}$	8	100	17	7	96	21	100	11	19	31
Tetratriacontane	$C_{34}H_{70}$		3			4		1			0.2
Pentatriacontane	$C_{35}H_{72}$		19			13		2			0.7
Hexatriacontane	$C_{36}H_{74}$					1					
Heptatriacontane	$C_{37}H_{76}$		2			3					
Biomarkers (hydrocarbons relative to n-alkanes, oxygenated relative presence)											
a-Amyrin	$C_{30}H_{50}O$	+			++				++		
ß-Amyrin	$C_{30}H_{50}O$	+			+++				+++		
a-Boswellic acid	$C_{30}H_{48}O_3$								+++++		
ß-Boswellic acid	$C_{30}H_{48}O_3$								+++++		
a- & ß-Amyrones	$C_{30}H_{48}O$	+++				++			+++		
FriedelinC30H50O	$C_{30}H_{50}O$	+		+++		+	++				++
Friedelanol	$C_{30}H_{52}O$			+		+++					
Friedelane	$C_{30}H_{52}$								7		
Olean- 12 - ene	$C_{30}H_{50}$	1				4			15		
Taraxerene	$C_{30}H_{50}$				1.4	19			40		
Dihydronyctanthic acid	$C_{30}H_{50}O_2$	+			+				+		
Dihydroroburic acid	$C_{30}H_{50}O_2$	+			+				+		
Dihydrocanaric acid	$C_{30}H_{50}O_2$	+			+				+		
Squalene	$C_{30}H_{50}$					3					

*Compound with the highest concentration is assigned a value of 100

Australia, Punta Arenas (S. America), Nigeria and the United States are all generally at C_{29} (Table 3). Thus the higher C_{max} of these samples reflects the higher ambient temperature which results in biosynthesis of longer carbon chain lipids which have higher melting points (Simoneit, 1977).

Since biosynthetic *n*-alkanes exhibit a strong odd-carbon number predominance, the concentration ratio of odd-to-even number homologues (carbon preference index, CPI, Mazurek and Simoneit, 1984) of plant wax *n*-alkanes will have values of >1. The CPI values of the *n*-alkane homologues obtained in this study range from 3.0-25.2. These values are comparable to the CPI values reported for other studies (Table 3).

n-Alkanoic and n-Alkenoic Acids

Another major compound class commonly identified in epicuticular plant waxes is the group of long-chained saturated *n*-fatty acids (Kolattukudy, 1970; Simoneit, 1977, 1989; Simoneit and Mazurek, 1982; Rieley *et al.* 1991). In this study, the *n*-alkanoic acids are found in both fractions 2 and 3 of the extracts. The n-alkanoic acids show a strong even-to-odd carbon number predominance with C_{max} at C_{16} and C_{20}, C_{24}, C_{28} or C_{32}. Of all the species investigated five species exhibit C_{max} at C_{16} and two have C_{max} at C_{28}, with the remainder at other carbon numbers (Table 1). The CPI_{even} values range from 5.9 to 97.5. For comparison, the CPI_{even} values reported for other studies range from 13.1 to 34.0 (Table 3).

Unsaturated *n*-fatty acids have also been identified and quantified in the leaf wax extracts. Oleic ($C_{18:1}$), linolenic ($C_{18:2}$), and linoleic ($C_{18:3}$) acids are the major unsaturated *n*-fatty acids (*n*-alkenoic acids) identified here, however, they occur as trace components.

n-Alkanols and n-Alkanals

Both *n*-alkanols and *n*-alkanals are synthesized enzymatically by plants from precursor *n*-alkanoic acids (Kolattukudy, 1970). The long-chain primary *n*-alkanols with even carbon number predominances and C_{max} at C_{26}, C_{28}, C_{30}, C_{32} or C_{34} have been identified in the present study. They range from C_{10} to C_{36} with CPI_{even} values from 5.3 to ∞ (Table 1). These values are in the same range as reported for other areas (Table 3). The *n*-alkanals are not as common in these samples. Only two samples had long chain alkanals as minor components of the wax.

Molecular Markers (Biomarkers)

Biogenic molecular markers (i.e. biomarkers) include the triterpenes and terpenoids or their derivatives such as phytosterols, triterpenols and triterpenol acids, sesquiterpenoids and diterpenoids.

(a) *Phytosterols*

In this study, the phytosterols are found in the alcohol fractions of the plant wax extracts, but only traces of mainly b-sitosterol were detected in some of the samples. Phytosterols are not major components of wax from tropical vegetation.

TABLE 3

A comparison of lipid characteristics of vascular palant wax from various global locations.

Locations	Latitude (approx.)	Mean ambient temperature (^{0}C)	Total extract (mg/g)	n-Alkanes			n-Alkanoc acids			n-Alkanols			Ref.
				Yield (g/g)	CPI	C^{max}	Yield (g/g)	CPI	C^{max}	Yield (g/g)	CPI	C^{max}	
Malaysia	3^0N	28	1.2-35.4	40-4600	3.0-25.5	29,31,33	n.a.	5.9-97.5	16,14,28, 32	n.a.	5.3-	16,18,26, 28,30,32, 34	Present study
Australia	34^0S	22	80	1400	16.0	29	2650	17.1	16.25	80000	47.5	26	Simoneit el al. (1991b)
Brazil (Amazonia)	3^0C	30	n.a.	n.a.	14.1	29	n.a	25.0	16,22	n.a.	8.5	30	Simoneits et. al. (1990)
Chile (Punta Arenas)	54^0S	10	630 mg	12 ng/g	30	29	n.a.	34.0	16,26	n.a.	10.0	22,24	Simoneits et. al. (1991a)
Nigeria	9^0N	24	n.a.	300	28.4	29	200	13.1	16,20,24 30	240	14.6	30	Simoneits et. al. (1988)
U.S.A (California)	39^0N	20	6	184	59	29	320	18.2	12,24,30	1680	18.4	16,24,30	Simoneits and Mazurek (1882)
Japan	34^0N	16	n.a.	100-3800	n.a.	29,31	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	Nishimoto (1974)
U.A.S. (Oregon)	45^0N	18	n.a.	n.a.	3.6-17	23-33	n.a.	2.3-	16-28	n.a.	3-11	22-28	Standley (1988);Chen (1992)

n.a. = not analyzed

(b) *Triterpenoids*
The alcohol fraction of *M. ferrea* wax contains a-boswellic and b-boswellic acids as major components, with minor amounts of a-amyrin and b-amyrin (Table 2). The mass spectrum of methyl b-boswellate trimethylsilyl ether ($C_{34}H_{58}O_3Si$, methyl olean-12-en-3a-ol-24-oate-3-trimethylsilyl ether) exhibits a molecular ion, M-CH_3, M-CH_3OH_2 and the characteristic key ions of the oleanene rings C-E at m/z 203 and 218. Both a-amyrin and b-amyrin are lesser constituents in the other samples (Table 2). The other triterpenoids found are also listed in Table 2. Friedelin and friedelanol are significant components of *E. grandis* and *H. brasiliensis* wax, respectively. *M. ferrea* wax contains various triterpenes such as the amyrins and minor oxidation and/or alteration products, such as amyrones, *seco*-amyrins (i.e. dihydroroburic and dihydronyctanthic acids) and *seco*-lupeol (i.e. dihydrocanaric acid). These latter compounds are also recognizable as minor components in some of the other waxes where the parent compounds are present. The mass spectra of these *seco* compounds are mainly the dihydro derivatives. Photo-oxidation of the precursors should initially yield the intermediates such as roburic, nyctanthic and canaric acids, however, the dihydro acids have been reported to form directly by photochemical cleavage of amyrones (Arigoni *et al.* 1960). Methyl dihydronyctanthate exhibits a small molecular ion and M-15 with the characteristic key ions of the oleanene rings C-E at m/z 203 and 218. The GC elution order is as for the amyrins, i.e. dihydronyctanthic acid before dihydroroburic acid.

(c) *Sesqui- and Diterpenoids*
No sesquiterpenoids nor diterpenoids were detected in the wax extracts of these samples.

Phthalates
The presence of dialkyl esters of benzene-1,2-dicarboxylic acid (phthalates) is significant (e.g. in fraction 4 of *E. grandis* and *L. indica*). These compounds are not known to be present in waxes of plants, but instead are ubiquitous plasticizers and do occur in urban atmospheres. Thus the presence of these compounds in wax extracts further substantiates the findings of Hosker and Lindberg (1982) and Shanley (1989) that leaf surfaces are an important sink for airborne particulate matter originating from anthropogenic sources.

CONCLUSION

The epicuticular waxes of the common tree species of the Klang Valley consist of the typical lipid fractions with minor biomarkers. *Mesua ferrea* had the highest wax yield and *Eugenia grandis* the lowest. Hentriacontane (C_{31}) was the dominant *n*-alkane for most species, followed by tritriacontane (C_{33}) and nonacosane (C_{29}), respectively. The *n*-alkanoic acids had a C_{max} at C_{16} and C_{20}, C_{24}, C_{28} or C_{32}. The *n*-alkanols had a C_{max} at C_{28} for most species and at C_{26}, C_{30}, C_{32} or C_{34} for the others. One distinct feature that is observed in this study is that the C_{max} for *n*-alkanes is higher than that reported for other locations. The biomarkers identified are b-sitosterol, a- and b-boswellic acids (as the major components), a- and b-amyrins, a- and b-amyrones, friedelin and friedelanol. Other trace biomarkers are friedelane, olean-12-ene, taraxerene, squalene, dihydronyctanthic acid, dihydroroburic acid and dihydrocanaric acid. These biomarkers are among the most common biomarkers identified in the atmosphere.

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