



Original Research Article

A New Phytosteroid and Aliphatic Constituents from the Roots of *Albizzia lebeck* Benth

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ABSTRACT

Albizzia lebeck Benth is found in south eastern Asia and Australia and has been introduced in central America, Colombia, Venezuela and Brazil. Its root bark is used to cure inflammation, blood related diseases, leucoderma, itching, skin diseases, piles and bronchitis. Phytochemical investigation of a methanolic extract of the roots of *A. lebeck* led to the isolation of a new phytosteroid characterized as stigmast-4,20(21),22-trien-3-one (lebbeksterone) (**2**), an aliphatic alcohol, *n*-tricontan-10 α -ol (isotriacontanol) (**1**), two fatty acid esters, *n*-tricosanyl *n*-octadec-9-en-1-oate (*n*-tricosanyl oleate) (**3**) and *n*-pentacosanyl *n*-octadec-9-en-1-oate (*n*-pentacosanyl oleate) (**4**) along with a known fatty acid docos-3-en-1-oic acid (docosenoic acid) (**5**). The structures of all these phytoconstituents have been elucidated on the basis of spectral data analysis and chemical reactions.

Keyword: *Albizzia lebeck*; Fabaceae; roots; stigmast-4, 20(21), 22-trien-3-one; *n*-tricontan-10 α -ol; fatty acid esters

INTRODUCTION

Albizzia lebeck Benth (Fabaceae) is a fast-growing, medium-sized deciduous tree with a spreading umbrella-shaped crown of thin foliage and smooth, fissured, greyish-brown bark. It is a native to deciduous and semi deciduous forests in eastern Pakistan,

throughout India, Nepal, Bangladesh, Myanmar, Indonesia, Sri Lanka, Thailand and Australia and has been introduced as an ornamental tree throughout the tropics and northern subtropics including the Greater and Lesser Antilles, Central America, Colombia, Venezuela and

Brazil [1]. Its root bark is used to cure inflammation, blood related diseases, leucoderma, itching, skin diseases, piles and bronchitis. The root bark powder is prescribed to strengthen the gums, when they are spongy and ulcerative [2-4]. The bark contained saponins [5]; the seeds yielded alkaloids, viz., budmunchiamine L1-L6 [6-8]. Two phytoconstituents (-)-2,3-cis-3,4-cis-3-O-methylmelaccacidin and 3'-O-methylmelanoxetin were isolated from heartwood [9]. The roots possessed echinocystic acid (saponin) glycoside, 2'- α -hydroxyoctyl hexadecanoate, salicylic acid-2-O- β -D-glucofuranosyl-6'-octadec-9"-enoate, docos-3-en-1-oic acid and fatty acid ester [10-12]. The present paper describes the isolation and characterization of a new steroid along with four aliphatic constituents from the roots of *A. lebbeck*.

MATERIALS AND METHODS

General Experimental Procedure

The melting points were determined on a Perfit apparatus and are uncorrected. The IR spectra were recorded in KBr pellet on Win IR FTS 135 instrument (Biorad, USA). The ^1H (300 MHz) and ^{13}C (75 MHz) NMR spectra were run by Bruker spectrosin NMR instrument in CDCl_3 , using TMS as an internal standard. The FAB MS were scanned at 70 eV on a Jeol D-300 instrument. Column chromatography was performed on silica gel (Merck, 60-120 mesh) and thin-layer chromatography on silica gel G coated TLC plates (Merck). Spots were visualized by exposure to iodine vapours, UV radiation and by spraying reagent.

Plant Material

The roots of *A. lebbeck* were collected from West Champaran, Bihar, (India) and identified by Dr. H. B. Singh, Scientist F and Head, Raw Materials, Herbarium and Museum, National Institute of Science Communication and Information Resources (NISCAIR), New Delhi,

India. A voucher specimen (No. NISCAIR/RHMD/Consult/-2008-09/1114/145) was deposited in the Herbarium of NISCAIR, New Delhi.

Extraction and Isolation

The roots (1.6 kg) were shade dried, coarsely powdered and extracted exhaustively with methanol as the reported procedure [13-16]. The methanolic extract was concentrated under reduced pressure to obtain a dark green viscous mass (85.5 g). For isolation of the compounds, the extract was dissolved in little amount of methanol and adsorbed on silica gel (60-120 mesh) for column for preparation of a slurry [14-16]. The slurry was air-dried and chromatographed over a silica gel column packed in petroleum ether. The column was eluted successively with petroleum ether, mixture of petroleum ether and chloroform (9:1, 3:1, 1:1 and 1:3), pure chloroform and finally the mixture of chloroform and methanol (99:1, 98:2, 96:4, 95:5, 97:3, 9:1). Various fractions were collected separately and matched by TLC to check homogeneity. Similar fractions (having the same R_f values) were combined and crystallized. The isolated compounds were recrystallized.

RESULTS

Isotriacontanol (1)

Elution of the column with petroleum ether-chloroform (3:1) yielded colorless crystals of **1**, recrystallized from chloroform-methanol (1:1), 150 mg (0.17% yield); R_f : 0.64 (CHCl_3 -MeOH, 9:1); m.p. 88-89 °C; UV λ_{max} (MeOH): 225 nm ($\log \epsilon$ 3.1); IR ν_{max} (KBr): 3446, 2923, 2854, 1641, 1462, 1112, 722 cm^{-1} . ^1H NMR (CDCl_3): δ 4.05 (1H, m, $w_{1/2}$ =6.6 Hz, H-10 β), 2.31 (2H, m, CH_2), 1.61 (6H, m, 3x CH_2), 1.25 (46H, brs, 23 x CH_2), 0.88 (3H, t, J =6.9 Hz, Me-1), 0.86 (3H, t, J =6.6 Hz, Me-30); ^{13}C NMR (CDCl_3): δ 74.11 (C-10), 32.06 (CH_2), 29.57 (21 x CH_2), 29.03 (CH_2), 29.01 (CH_2), 28.99 (CH_2), 26.13 (CH_2), 22.55 (CH_2), 14.17 (2 x Me); +ve ion FAB MS m/z (*rel. int.*): 438 [M^+ ($\text{C}_{30}\text{H}_{62}\text{O}$) (26.3), 423 (55.7), 420

(36.1), 408 (72.2), 311(16.3), 293 (17.5), 281 (78.6), 157 (31.8), 127 (23.4).

Lebbeksterone (2)

Elution of the column with petroleum ether-chloroform (1:3) afforded colorless crystals of **2**, recrystallized from acetone, 710 mg (0.81% yield); R_f : 0.52 (CHCl₃); m.p. 252-253 °C; UV λ_{max} (MeOH): 223 nm (log ϵ 5.5); IR ν_{max} (KBr): 2925, 2845, 1705, 1645, 1475, 1360, 1290, 1015 cm⁻¹; ¹H NMR (CDCl₃): δ 5.53 (1H, s, H-4), 5.12 (1H, d, $J=6.3$ Hz, H-22), 5.06 (1H, dd, $J=6.3, 6.1$ Hz, H-23), 5.03 (1H, s, H₂-21a), 4.99 (1H, s, H₂-21b), 1.02 (3H, s, Me-19), 0.87 (3H, d, $J=6.2$ Hz, Me-26), 0.84 (3H, d, $J=6.5$ Hz, Me-27), 0.81 (3H, d, $J=6.5$ Hz, Me-29), 0.65 (3H, s, Me-18), 2.77-1.10 (24H, m, 9 x CH₂, 6 x CH); ¹³C NMR (CDCl₃): δ 38.19 (C-1), 31.95 (C-2), 212.11 (C-3), 129.52 (C-4), 139.55 (C-5), 19.01 (C-6), 34.42 (C-7), 30.06 (C-8), 51.26 (C-9), 39.33 (C-10), 21.71 (C-11), 40.07 (C-12), 41.27 (C-13), 55.85 (C-14), 24.66 (C-15), 28.53 (C-16), 55.03 (C-17), 11.14 (C-18), 21.14 (C-19), 138.11 (C-20), 117.01 (C-21), 130.04 (C-22), 129.58 (C-23), 48.04 (C-24), 29.40 (C-25), 23.04 (C-26), 21.40 (C-27), 28.53 (C-28), 12.11(C-29); +ve ion FAB MS m/z (*rel. int.*): 408 [M]⁺ (C₂₉H₄₄O) (27.1), 293 (21.6), 272 (32.8), 271 (18.2), 256 (19.9), 241 (15.7), 232 (14.2), 229 (13.1), 213 (21.6), 190 (20.3), 176 (19.3), 175 (18.1), 162 (26.7), 160 (3.1), 148 (31.2), 147 (41.3), 145 (11.5), 137 (21.5), 136 (38.5), 134 (40.6), 133 (34.2), 122 (49.9), 119 (52.1), 108 (75.8), 85 (56.1), 43 (33.1).

n-Tricosanyl oleate (3)

Elution of the column with chloroform furnished colourless crystals of **3**, recrystallized from chloroform-methanol (1:1), 210 mg (0.25% yield); R_f : 0.69 (CHCl₃); m.p. 79-80 °C; UV λ_{max} (MeOH): 225 nm (log ϵ 3.7); IR ν_{max} (KBr): 2922, 2854, 1745, 1638, 1461, 1373, 1165, 723 cm⁻¹; ¹H NMR (CDCl₃): δ 5.35 (2H, m, H-9, H-10), 4.28 (1H, t, $J=6.6$ Hz, H₂-1'), 2.31 (2H, t, $J=7.2$ Hz, H₂-2), 2.04 (2H, m, H₂-8), 2.01 (2H, m, H₂-11), 1.64 (6H, brs, 3 x CH₂), 1.25 (58H, brs, 29 x CH₂),

0.88 (3H, t, $J=6.5$ Hz, Me-18), 0.84 (3H, t, $J=6.3$ Hz, Me-23'); ¹³C NMR (CDCl₃): δ 173.26 (C-1), 129.98 (C-9), 128.04 (C-10), 62.07 (C-1'), 34.02 (CH₂), 31.90 (CH₂), 31.50 (CH₂), 29.67 (27 x CH₂), 29.31(CH₂), 27.17(CH₂), 25.60 (CH₂), 24.84 (CH₂), 22.66 (CH₂), 14.08 (CH₃-18), 14.04 (CH₃-23'); +ve ion FAB MS m/z (*rel. int.*): 604 [M]⁺(C₄₁H₈₀O₂) (28.3), 339 (100), 281 (13.5), 265 (41.9), 139 (11.8), 113 (15.1).

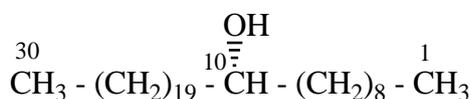
n-Pentacosanyl oleate (4)

Further elution of the column with chloroform furnished colorless crystals of **4**, recrystallized from chloroform-methanol (1:1), 270 mg (0.31 % yield); R_f : 0.56 (CHCl₃); m.p. 86-90 °C; UV λ_{max} (MeOH): 223 nm (log ϵ 2.9); IR ν_{max} (KBr): 2923, 2854, 1745, 1639, 1460, 1374, 1164, 723 cm⁻¹; ¹H NMR (CDCl₃): δ 5.36 (1H, m, H-9), 5.34 (1H, m, H-10), 4.12 (2H, brs, H₂-1'), 2.34 (1H, t, $J=7.2$ Hz, H₂-2), 2.06 (2H, m, H₂-8), 2.02 (2H, m, H₂-11), 1.78 (2H, m, CH₂), 1.54 (4H, m, 2 x CH₂), 1.30 (2H, brs, CH₂), 1.04 (2H, m, CH₂), 0.88 (3H, t, $J=6.6$ Hz, Me-18), 0.83 (3H, t, $J=7.8$ Hz, Me-25'); ¹³C NMR (CDCl₃): δ 172.16 (C-1), 131.54 (C-9), 129.15 (C-10), 62.18 (C-1'), 34.55 (CH₂), 34.49 (CH₂), 31.95 (CH₂), 31.04 (CH₂), 30.47 (CH₂), 29.73 (2 x CH₂), 29.73 (CH₂), 29.25 (CH₂), 27.22 (CH₂), 25.64 (CH₂), 24.89 (CH₂), 22.31(CH₂), 21.16 (CH₂), 19.01 (CH₂), 13.03 (Me-18), 13.01(Me-25); +ve ion FAB MS m/z (*rel. int.*): 632 [M]⁺ (C₄₃H₈₄O₂) (11.3), 395 (31.9), 367 (32.8), 265 (23.7), 237 (11.5).

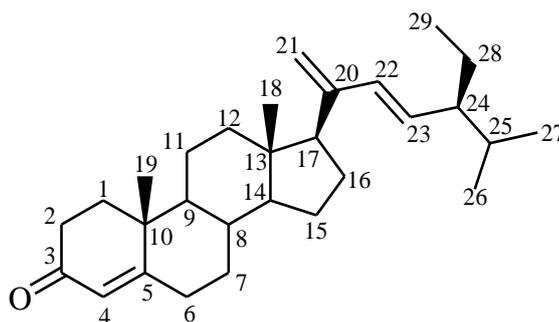
n-Docosenoic acid (5)

Elution of the column with chloroform-methanol (99:1) afforded colourless crystals of **5**, recrystallized from acetone, 1130 mg (1.32% yield); R_f : 0.72 (CHCl₃-MeOH, 4:1); m.p. 86-87 °C; UV λ_{max} (MeOH): 225 nm (log ϵ 3.7); IR ν_{max} (KBr): 3340, 1708, 1630, 724 cm⁻¹; ¹H NMR (CDCl₃): δ 5.27 (2H, m, H-3, H-4), 2.29 (2H, t, $J=7.5$ Hz, H₂-2), 1.96-1.53 (6H, m, 3 x CH₂), 1.55 (2H, m, CH₂), 1.53 (2H, m, CH₂), 1.18 (32H, brs, 16 x CH₂), 0.80 (3H, t, $J=6.9$ Hz, Me-22); ¹³C NMR (CDCl₃): δ 176.65 (C-1), 129.97(C-3), 128.03 (C-

4), 34.01-22.66 (18 x CH₂), 14.08 (CH₃-22); +ve ion FAB MS *m/z* (*rel. int.*): 338 [M]⁺ (C₂₂H₄₂O₂) (32.9).



Isotriacontanol (**1**)



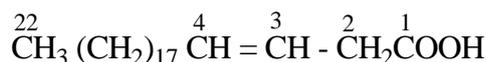
Lebbeksterone (**2**)



n-tricosanyl oleate (**3**)



n-pentacosanyl oleate (**4**)



n-Docosenoic acid (**5**)

Fig 1. The structures of compounds 1-5

Compounds **1** and **2** were obtained as colourless crystals from petroleum ether-chloroform (3:1) and (1:3) mixtures, respectively. Compound **3** and **4** were eluted with chloroform eluants from the column as colourless crystalline products. Compound **5** was obtained as a colourless mass from chloroform-methanol (99:1) eluants. Compound **1** did not respond positively with any decolorizing reagents and other specific chemical tests of compounds suggesting

saturated nature of the molecule. Compound **2** gave Liebermann-Burchard test positive indicating steroidal nature of the molecule. Compounds **3** and **4** decolorized bromine water supporting unsaturated nature of the molecules. Compound **5** yielded effervescence with sodium bicarbonate solution indicating carboxylic nature of the molecule.

DISCUSSION

Compound **1**, named isotriacontanol, showed IR absorption bands for hydroxyl group (3446 cm^{-1}) and long aliphatic chain (722 cm^{-1}). The mass spectrum of **1** exhibited a molecular ion peak at m/z 438 corresponding to the molecular formula of a saturated aliphatic alcohol, $\text{C}_{30}\text{H}_{62}\text{O}$. The mass spectrum displayed C_nH_{2n} and $\text{C}_n\text{H}_{2n-1}$ fragmentation peaks and most of the fragments were separated by 14 mass units that indicated straight chain nature of **1**. The prominent ion peaks generated at m/z 127 $[\text{CH}_3(\text{CH}_2)_8]^+$ and 311 $[\text{CH}_3(\text{CH}_2)_{19}\text{CHOH}]^+$ due to $\text{C}_9\text{-C}_{10}$ fission and 157 $[\text{CH}_3(\text{CH}_2)_8\text{CHOH}]^+$ and 281 $[\text{CH}_3(\text{CH}_2)_8]^+$ due to $\text{C}_{10}\text{-C}_{11}$ fission suggested the location of the hydroxyl group at C_{10} position. The ion peaks at m/z 420 $[\text{M-H}_2\text{O}]^+$ and 293 $[311-\text{H}_2\text{O}]^+$ supported the presence of hydroxyl group in the compound **1**. The ^1H NMR spectrum of **1** showed a one-proton broad multiplet at δ 4.05 with half width of 6.6 Hz assigned to β -oriented H-10 carbinol proton. Three signals as multiplets at δ 2.31 (2H) and 1.61 (6H) and a broad singlet at δ 1.25 (46H) were ascribed to the methylene protons. Two three-proton triplets at 0.88 ($J=6.9$ Hz) and 0.86 ($J=6.6$ Hz) were attributed to C-1 and C-30 primary methyl protons, respectively. The ^{13}C NMR spectrum of **1** exhibited important signals for carbinol carbon C-10 at δ 74.11, methylene carbons between δ 32.06 to 22.25 and methyl carbons at δ 14.17. The absence of any signal beyond δ 4.05 in the ^1H NMR spectrum and δ 74.11 in the ^{13}C NMR spectrum supported the saturated nature of **1**. On the basis of the foregoing discussion the structure of **1** has been elucidated as *n*-tricontan-10 α -ol (Fig 1).

Compound **2**, designated as lebbeksterone, responded positively to Liebermann-Buchardt test for steroids. Its IR spectrum showed characteristic absorption bands for carbonyl group (1705 cm^{-1}) and unsaturation (1645 cm^{-1}). The +ve FAB mass spectrum displayed a molecular ion peak at m/z 408 consistent with a steroidal molecular formula, $\text{C}_{29}\text{H}_{44}\text{O}$ which was

supported by ^{13}C NMR spectrum. It indicated the presence of eight double bond equivalents; four of them were adjusted in the steroidal carbon framework, three in olefinic linkages and one in a carbonyl group. The diagnostically important peaks were observed at m/z 108 $[\text{C}_{5,6}\text{-C}_{9,10}\text{ fission}]^+$, 122 $[\text{C}_{6,7}\text{-C}_{9,10}\text{ fission}]^+$, 272 and 136 $[\text{C}_{7,8}\text{-C}_{9,10}\text{ fission}]^+$, that indicated the saturated nature of ring B and the presence of vinylic linkage in ring A. The ion peaks at m/z 162 $[\text{C}_{8,14}\text{-C}_{9,11}\text{ fission}]^+$, 176 $[\text{C}_{8,14}\text{-C}_{11,12}\text{ fission}]^+$ and 190 $[\text{C}_{8,14}\text{-C}_{12,13}\text{ fission}]^+$ supported the saturated nature of ring C. The other important ion fragments at m/z 137 $[\text{C}_{10,17},\text{ side chain}]^+$, 271 $[\text{C}_{17,20}\text{ fission, M-side chain}]^+$, 229 $[\text{271-ring D}]^+$ and 256 $[\text{271-Me}]^+$ suggested the presence of saturated ring D and a C-10 unsaturated side chain. The ion peaks at m/z 134 $[\text{162-CO}]^+$, 148 $[\text{176-CO}]^+$, 162 $[\text{190-CO}]^+$ and 213 $[\text{241-CO}]^+$ suggested the presence of carbonyl group in ring A which was placed at C-3 on biogenetic considerations. The ion fragments at m/z 43 $[\text{C}_{24}\text{-C}_{25}\text{ fission}]^+$, and 85 $[\text{C}_{23}\text{-C}_{24}\text{ fission}]^+$ indicated the presence of exocyclic vinylic linkage at C-20 and a double bond at C-22 and an ethyl group in the side chain at C-24. The ^1H NMR spectrum of **2** exhibited three one-proton downfield broad singlets at δ 5.53, 5.03 and 4.99 assigned correspondingly to H-4, H-21a and H-21b vinylic protons. A doublet and a double-doublet at δ 5.12 ($J=6.3$ Hz) and 5.06 ($J=6.3, 6.1$ Hz), each integrating for one proton, were ascribed correspondingly to H-22 and H-23 vinylic protons. Two doublets, three-protons each, at δ 0.87 ($J=6.2$ Hz) and 0.84 ($J=6.5$ Hz), were assigned to C-26 and C-27 secondary methyl protons. The tertiary methyl protons resonated as two three-proton broad singlets at δ 1.02 (Me-19) and 0.65 (Me-18). The appearance of methyl protons in the region δ 1.02-0.65 suggested that these groups were attached to saturated carbons. The ^{13}C NMR spectrum of **2** displayed signals for carbonyl carbon at δ 212.11 (C-3) and vinylic carbons appeared at δ 129.52 (C-4), 139.55 (C-5), 138.11

(C-20), 117.01(C-21), 130.04 (C-22) and 129.58 (C-23). The ^{13}C NMR values of **2** were compared with the reported data of steroidal constituents [13, 14]. On the basis of above discussion the structure of **2** was formulated as stigmast-4, 20 (21), 22-trien-3-one (Fig 1). This is a new ketosteroid isolated from a plant source.

Compound **3**, *n*-tricosanyl oleate, showed IR absorption bands for ester group (1745 cm^{-1}), unsaturation (1638 cm^{-1}) and long aliphatic chain (723 cm^{-1}). Its mass spectrum displayed a molecular ion peak at m/z 604 corresponding to an aliphatic ester, $\text{C}_{41}\text{H}_{80}\text{O}_2$. A base peaks at m/z 339 $[\text{O}(\text{CH}_2)_{22}\text{CH}_3]^+$, along with the fragment ion peaks at m/z 265 $[\text{CH}_3(\text{CH}_2)_7\text{CH}=\text{CH}-(\text{CH}_2)_7\text{CO}]^+$ and 281 $[\text{CH}_3(\text{CH}_2)_7\text{CH}=\text{CH}(\text{CH}_2)_7\text{COO}]^+$ aroused due to ester linkage fission suggesting that **3** was a C_{18} fatty acid esterified with the C_{23} -alcohol. Other fragment ion peaks at m/z 113 $[\text{C}_{10}\text{-C}_{11}\text{ fission, CH}_3(\text{CH}_2)_7]^+$ and 139 $[\text{C}_8\text{-C}_9\text{ fission, CH}_3(\text{CH}_2)_7\text{CH}=\text{CH}]^+$ supported the presence of vinylic linkage at C_9 . Most of the ion fragments were separated by a mass units difference of 14 indicating the presence of unbranched aliphatic chain in the compound. The ^1H NMR spectrum of **3** displayed a two-proton multiplet at δ 5.35 assigned to vinylic protons H-9 and H-10. A two-proton triplet at δ 4.28 ($J=6.6\text{ Hz}$) was ascribed to oxygenated methylene protons $\text{H}_2\text{-1}'$. A two-proton triplet at δ 2.31 ($J=7.2\text{ Hz}$) was assigned to $\text{H}_2\text{-2}$ methylene protons adjacent to the ester group. Two two-proton multiples at δ 2.04 and 2.01 were assigned correspondingly to $\text{H}_2\text{-8}$ and $\text{H}_2\text{-11}$ methylene protons adjacent to vinylic linkage. The remaining methylene protons appeared as broad singlets at δ 1.64 (6H) and 1.25 (58H). Two three-proton triplets at δ 0.88 ($J=6.5\text{ Hz}$) and 0.84 ($J=6.3\text{ Hz}$) were ascribed to primary Me-18 and Me-23' methyl protons, respectively. The ^{13}C NMR spectrum of **3** exhibited important signals for ester carbon at δ 173.26 (C-1), vinylic carbons at δ 129.98 (C-9) and 128.04 (C-10); oxygenated methylene carbon δ 62.07 (C-1') and primary methyl

carbons at δ 14.08 ($\text{CH}_3\text{-18}$), 14.04 ($\text{CH}_3\text{-23}'$). The remaining methylene carbons resonated in the range between δ 34.02 to 22.66. The hydrolysis of **3** yielded oleic acid (co-TLC comparable). On the basis of above discussion the structure of **3** was characterized as *n*-tricosanyl *n*-octadec-9-en-1-oate (Fig 1).

Compound **4**, *n*-pentacosanyl oleate, showed characteristic IR absorption bands for ester group (1745 cm^{-1}), unsaturation (1639 cm^{-1}) and long aliphatic chain (723 cm^{-1}). Its +ve FAB mass spectrum displayed a molecular ion peak at m/z 632 corresponding to molecular formula of an aliphatic ester $\text{C}_{43}\text{H}_{84}\text{O}_2$. It indicated the presence of two-double bond equivalents that were adjusted one each in the vinylic linkage and ester group. The fragment ion peaks arising at m/z 395 $[\text{CH}_3(\text{CH}_2)_{24}\text{OCO}]^+$, 367 $[\text{CH}_3(\text{CH}_2)_{24}\text{O}]^+$, 265 $[\text{CH}_3(\text{CH}_2)_7\text{-CH}=\text{CH}(\text{CH}_2)_7\text{CO}]^+$ and 237 $[\text{CH}_3(\text{CH}_2)_7\text{CH}=\text{CH}(\text{CH}_2)_7]^+$ due to ester linkage fission suggested that **4** was an ester of an unsaturated C_{18} fatty acid esterified with the C_{25} -alcohol. Most of the fragments were separated by 14 mass units indicating the straight chain nature of the compound. The ^1H NMR spectrum of **4** displayed two downfield multiplets, each integrating for one-proton, at δ 5.36 and 5.34 assigned correspondingly to H-9, H-10 vinylic protons. A two-proton triplet at δ 2.34 was ascribed to $\text{H}_2\text{-2}$ methylene protons adjacent to the ester group. An unresolved broad signal appeared at δ 4.12 integrating for two protons, was ascribed to $\text{H}_2\text{-1}'$ oxygenated methylene protons. The remaining methylene protons resonated between δ 2.06 to 1.04. Two three-proton triplets at δ 0.88 ($J=6.6\text{ Hz}$) and 0.83 ($J=7.8\text{ Hz}$), were assigned correspondingly to Me-18 and Me-25' primary methyl protons. The ^{13}C NMR spectrum of **4** displayed important signals for ester carbon at δ 172.61 (C-1); vinylic carbons at δ 131.54 (C-9) and 129.15 (C-10); methylene carbons between δ 34.55 to 19.01 and primary methyl carbons at δ 13.03 (Me-18) and 13.01 (Me-25'), respectively. The

hydrolysis of **4** yielded oleic acid (co-TLC comparable). On the basis of above discussion, the structure of **4** has been elucidated as *n*-pentacosanyl *n*-octadec-9-en-1-oate (Fig 1). Compound **5** was the known fatty acid identified as *n*-docosenoic acid (Fig 1).

CONCLUSION

Phytochemical investigation of the roots of *Albizia lebbek* led to the isolation and characterization of one each of aliphatic secondary alcohol, steroidal ketone and fatty acid and two fatty ester for the first time. This study has enhanced the phytochemical nature of the plant. These compounds may be used as chromatographic markers for standardization.

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CONFLICT OF INTEREST STATEMENT

The authors declare that they have no competing interests.

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