BENTHAM OPEN

CrossMark

The Open Plant Science Journal

Content list available at: www.benthamopen.com/TOPSJ/

DOI: 10.2174/1874294701710010082

RESEARCH ARTICLE

Chemical Constituents from the Rhizomes of *Cyperus Rotundus* L.

Shahnaz Sultana[1, 2], Mohammed Ali[1,*] and Showkat Rasool Mir[1]

[1]*Phytochemistry Research Laboratory, Faculty of Pharmacy, Jamia Hamdard, New Delhi 110 062, India .*
[2]*College of Pharmacy, Jazan University, Jazan, Saudi Arabia.*

Received: February 28, 2017 Revised: May 11, 2017 Accepted: May 15, 2017

Abstract:

Background:

Cyperus rotundus L. (Cyperaceae), is a perennial sedge distributed throughout India and other parts of the world. Its tubers are used as an appetizer, febrifuge and to treat bleeding, blisters, boils, cough, diarrhea, inflammation, lacteal disorders, rheumatoid arthritis, stomach ailments, skin rashes, thirst, vomiting, worm infestation and wounds.

Objective:

Our study was planned to isolate chemical constituents from the rhizomes of *C. rotundus* and to characterized their structures.

Method:

The air-dried rhizome powder was exhaustively extracted with methanol in a Soxhlet apparatus. The concentrated methanol extract was adsorbed on silica gel (60-120 mesh) for the preparation of a slurry. The dried slurry was chromatographed over silica gel column packed in petroleum ether. The column was eluted with petroleum ether, chloroform and methanol, successively, in order of increasing polarity to isolate the compounds.

Results:

Phytochemical investigation of the tubers led to isolate a sesquiterpenone characterized as 12-methyl cyprot-3-en-2-one-13-oic acid (**1**), two aliphatic ketone viz. *n*-dotriacontan-15-one (**2**) and *n*-tetracontan-7-one (**8**), fatty esters *n*-pentadecanyl octadec-9, 12-dienoate (*n*-pentadecanyl linoleate, **3**), *n*-hexadecanyl linoleate (**4**), *n*-hexadecanyl oleate (**5**) and *n*-pentacos-13′-enyl octadec-9-enoate (*n*-pentacos-13′-enyl oleate, **9**), two steroidal esters **s**tigmast-5,22–dien-3*β* –olyl *n*-dodecanoate (stigmasterol laurate, **6**) and stigmast-5, 22-dien-3*β*-olyl *n*-tetradecanoate (stigmasterol myristate, **7**), β-sitosterol-3β-O-glucoside **(10)** and a triterpenic glycosidic ester lup-12, 20 (29)-dien-3β-ol-3-α-L-arabinopyranosyl-2'-oleate (lupenyl 3β-O-arabinpyranosyl 2′-oleate, **11**). The structures of these compounds were established by spectral data analysis and chemical reactions.

Conclusion:

A sesquiterpene identified as cyprot-3-en-2-one-14-oic acid, two aliphatic ketones, fatty esters, two steroidal esters, β-sitosterol-3β-O-glucoside and lupenyl 3β-O-arabinpyranosyl 2′-oleate were isolated for the first time from the rhizomes.

Keywords: *Cyperus rotundus*, Tubers, Acyl esters, Aliphatic ketones, Sterol esters, Lupenyl, Glycoside, Structure elucidation.

1. INTRODUCTION

Cyperus rotundus L. (Cyperaceae), syn. *C. maritimus* Bojer; *Pycreus rotundus* (L.) Hayek (Cyperaceae), known as nagarmotha, saad kufi and nut grass, is considered as one of the world's worst weeds. It is an indigenous to India, but now found in tropical, subtropical and temperate regions of the world [1]. It is a smooth , erect and perennial herb

* Address correspondence to this author at Phytochemistry Research Laboratory, Faculty of Pharmacy, Jamia Hamdard, New Delhi 110062, India; Tel: +9968281082; E-mail: maliphyto@gmail.com.

having wiry, slender, scaly, creeping, dark and persistent rhizomes [2]. Its tubers are used to treat loss of appetite, excess bleeding, blisters, boils, cough, diarrhea, fevers, inflammation, lacteal disorders, rheumatoid arthritis, stomach ailments, skin rashes, excessive thirst, vomiting, worm infestation and wounds [3 - 5]. In Ayurvedic medicine, the rhizomes are considered as an analgesic, antiseptic, antispasmodic, antitussive, aromatic, astringent, carminative, diaphoretic, diuretic, emmenagogue, litholytic, sedative, stimulant, stomachic, vermifuge and tonic; prescribed to treat amenorrhea, bronchitis, cervical cancer, colic, cough, diarrhea, dysentery, dysmenorrhea, dyspepsia, dysuria, fever, flatulence, food toxicity, indigestion, infertility, insect bites, intestinal parasites, deficient lactation, malaria, loss of memory, menstrual disorders, nausea, renal and vesical calculi, skin diseases, urinary tenesmus, vomiting and wounds [6, 7]. A decoction of the rhizomes with stem bits of *Tinospora cordifolia* and dried ginger is given to alleviate malarial fever. The rhizome decoction with the leaves of *Fumaria indica*, *Swertia chirayita*, black pepper and ginger is taken to relieve typhoid fever. The rhizome juice is given to allay constipation [2]. The rhizome mixed with ginger and honey is ingested against dysentery and gastric and intestinal troubles. A fresh tuber paste is applied to the breast as a galactagogue [8]. In the Egyptian folk medicine, the tubers are used as an anthelmintic, aphrodisiac, diuretic, sedative, carminative, stimulant, tonic, stomachic and as a remedy for renal colic and dysentery [9]. An essential oil from the tuber is used in perfumery and to make soap and insect repellent creams [7]. A decoction of the roots and tubers is an excellent antidote to all poisons. The tubers improve blood circulation and are effective in gynecological diseases caused by blood stagnation [10].

The rhizomes contained cyperene, cyperone, nor-rotundone, isorotundone, cypera-2,4(15)-diene, cyperadione and other essential oil components [11 - 19], sugetriol triacetate [20], caryophyllene, its oxide and caryophylla-6-one [21], patchoulenone, 4,7-dimethyl-1-tetralone and 10,12-peroxycalamenene [22], 4,5-secoeudesmanolide, 10-epi-4,5-secoeudesmanolide, cyclic acetal cyperolone, musktakone, nootkatone, rotunols [23], β-sitosterol, oleonolic acid-3-O-neohesperidoside [24], rhamnetin 3-O-rhamnosyl rhamnopyranoside [24], rotundines [25], flavonoids [26 - 28], phenylpropanoids [29 - 32] phenolic acids [30, 33], alkaloids [25], saponins [24, 33] and triterpenic glycosides [34]. The present paper describes the isolation and characterization of a sesquiterpenic keto acid, aliphatic ketones, fatty esters, steroids and a lupenyl glycosidic ester from the tubers of *C. rotundus* collected from Delhi.

2. MATERIAL AND METHODS

2.1. General Procedures

Melting points were determined on a Perfit melting point apparatus and are uncorrected. UV spectra were determined on Shimadzu-120 double beam spectrophotometer with methanol as a solvent. IR spectra were recorded in KBr pellet on Shimadzu FTIR-8400 spectrophotometer. The ^{1}H and ^{13}C-NMR spectra were scanned on Bruker DRX (300 MHz) instrument using TMS as an internal standard and coupling constants (J values) are expressed in Hertz (Hz). Mass spectra were recorded by affecting electron impact ionization at 70 eV on a Jeol SX-102 mass spectrometer equipped with direct inlet prob system. The *m/z* values of the more intense peaks are mentioned and the figures in bracket attached to each *m/z* values indicated relative intensities with respect to the base peak. Column chromatography was performed on silica gel (60-120 mesh; Qualigen, Mumbai, India). TLC was run on silica gel G 60 F_{254} precoated TLC plates (Merck, Mumbai, India). Spots were visualised by exposing to iodine vapours, UV radiations (254 and 366 nm) and spraying with ceric sulphate solution.

2.2. Plant Material

The tubers of *C. rotundus* were obtained from a Delhi market and authenticated by Prof. M.P. Sharma, Taxonomist, Department of Botany, Jamia Hamdard, New Delhi. A voucher specimen is preserved in the herbarium of the Department of Pharmacognosy and Phytochemistry, Jamia Hamdard, New Delhi.

2.3. Extraction and Isolation

The tubers (2.5 kg) were air dried, coarsely powdered and exhaustively extracted with methanol in a Soxhlet apparatus for 72 hours. The methanolic extract was evaporated under reduced pressure to get a brown viscous mass (186 g, 7.4% yield). The dried extract was dissolved in minimum quantity of methanol and added to silica gel (60-120 mesh) to prepare a slurry. It was air-dried, powdered and loaded on a silica gel column prepared in petroleum ether. The column was run with petroleum ether (b. p. 60 - 80°C), petroleum ether - chloroform (9:1, 3:1, 1:1, 1:3, *v/v*), chloroform and chloroform - methanol (99:1, 49:1, 19:5, 9:1, 17:3,4:1 7:3, 1:1, *v/v*). 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 to get pure compounds. The following compounds were isolated:

2.4. 12-Methyl Cyprot-3-en-2-one-13-oic Acid (1)

Elution of the column with petroleum ether gave colorless crystals of **1**, 78 mg, m. p. 55 - 56° C, UV λmax (MeOH): 213 nm (log ε 4.2); IR γ_{max} (KBr): 3363, 2927, 2842, 1703, 1685, 1645, 1426, 1372, 1218, 1178,1015, 982, 808 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 5.72 (1H, s, H-3), 2.59 (1H, m, $w_{1/2}$ = 8.7 Hz, H-1β), 2.05 (1H, m, H-12), 1.80 (1H, m, H_2-5β), 1.75 (1H, m, H_2-5α), 1.62 (1H, m, H-6α), 1.25 (3H, s, Me-9), 1.21 (3H, s, Me-10), 1.14 (3H, d, *J* = 6.2 Hz, Me-14), 0.83 (3H, s, Me-15), 0.65 (3H, d, *J* = 6.5 Hz, Me-11); ^{13}C NMR ($CDCl_3$): δ 58.68 (C-1), 210.98 (C-2), 133.16 (C-3), 145.66 (C-4), 31.98 (C-5), 32.76 (C-6), 45.29 (C-7), 41.21 (C-8), 24.83 (C-9), 26.98 (C-10), 19.57 (C-11), 29.76 (C-12), 181.93 (C-13), 16.88 (C-14), 8.41 (C-15); ESI MS *m/z* (rel.int.): 250 $[M]^+$ ($C_{15}H_{22}O_3$) (2.6).

2.5. *n*-Dotriacontan-16-one (2)

Elution of the column with petroleum ether – chloroform (9:1) furnished colorless powder of **2**, 188 mg, m. p. 54 – 56 °C, UV λmax (MeOH): 213 nm; IR γ_{max} (KBr): 2927, 2847, 1708, 1635, 1457, 1372, 1248, 1176, 1061, 723 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 2.34 (2H, m, H_2-14), 2.16 (2 H, m, H_2-16), 1.73 (2 H, m, CH_2), 1.65 (2 H, m, CH_2), 1.33 (2 H, m, CH_2), 1.28 (6 H, br s, 3 x CH_2), 1.23 (42 H, br s, 21 x CH_2), 0.86 (3 H, t, J = 6.6 Hz, Me-32), 0.83 (3 H, t, *J* = 6.1 Hz, Me-1); ^{13}C NMR ($CDCl_3$): δ 193.76 (C-15), 31.98 (CH_2), 29.78 (26 x CH_2), 29.37 (CH_2), 22.79 (CH_2), 14.89 (Me-1), 14.09 (Me-32); ESI MS *m/z* (rel. int.): 464 $[M]^+$ ($C_{32}H_{64}O$) (1.2), 225 (9.3), 177 (6.8).

2.6. *n*-Pentadecanyl Linoleate (3)

Elution of the column with petroleum ether - chloroform (3: 1) yielded colorless crystals of **3**, recrystallized from acetone – methanol (1:1); 132 mg; R_f: 0.70 (petroleum ether – chloroform - methanol, 2:7:1); m. p. 80-81 C; IR γ_{max} (KBr): 2917, 2849, 1737, 1641, 1462, 1261, 1098, 1022, 802, 719 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 5.82 (1H, m, H-10), 5.34 (1H, m, H-12), 5.01 (1H, m, H-9), 4.95 (1H, m, H-13),4.07 (2H, t, J = 6.6 Hz, H_2-1′), 2.48 (2H, m, H_2-11), 2.31 (2H, t, *J* = 7.2 Hz, H_2-2), 2.04 (2H, m, H_2-8), 2.02 (2H, m, H_2-14), 1.61 (4H, m, 2 x CH_2), 1.25 (38H, brs, 19 x CH_2), 0.89 (3H, t, *J* = 5.7 Hz, Me-18), 0.85 (3H, t, *J* = 6.3 Hz, Me-15′). ^{13}C NMR ($CDCl_3$): δ 169.83 (C-1), 139.17 (C-10), 135.76 (C-12), 119.81 (C-9), 116.89 (C-10), 64.13 (C-1′), 56.03 (CH_2), 42.83 (CH_2), 31.09 (CH_2), 29.70 (21 x CH_2), 22.68 (CH_2), 15.03 (Me-18), 14.16 (Me-15'). ESI MS *m/z* (rel.int.): 490 $[M]^+$ ($C_{33}H_{62}O_2$) (1.3), 279 (5.8), 263 (12.3), 227 (31.0).

2.7. *n*-Hexadecanyl Linoleate (4)

Further elution of the column with petroleum ether – chloroform (3:1) produced pale yellow semisolid mass of **4**, 182 mg, UV λmax (MeOH): 215 nm; IR γ_{max} (KBr): 2931, 2841, 1733, 1645, 1459, 1369, 1257, 1170, 891, 725 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 5.36 (1H, m, H-9), 5.33 (2H, m, H-10, H-12), 5.29 (1H, m, H-13), 4.12 (2H, t, *J* = 6.8 Hz, H_2-1′), 2.57 (2H, m, H-11), 2.35 (2H, t, *J* = 7.2 Hz, H_2-2), 2.06 (2 H, m, H_2-8), 1.73 (2 H, m, CH_2-14), 1.62 (2 H, m, CH_2), 1.55 (2H, m, CH_2), 1.34 (6 H, brs, 3 x CH_2), 1.28 (34 H, br s, 17 x CH_2), 0.87 (3 H, t, *J* = 6.2 Hz, Me-18), 0.84 (3 H, t, *J* = 6.5 Hz, Me-16′); ^{13}C NMR ($CDCl_3$): δ 172.96 (C-1), 133.28 (C-9), 128.27 (C-10), 125.91 (C-12), 118.08 (C-13), 64.47 (C-1′), 33.45 (CH_2), 32.21 (CH_2), 31.89 (CH_2), 29.75 (20 x CH_2), 29.64 (CH_2), 29.37 (CH_2), 22.67 (CH_2), 14.27 (Me-18), 14.13 (Me-16′); ESI-MS *m/z* (rel. int.): 504 $[M]^+$ ($C_{34}H_{64}O_2$) (1.2), 279 (11.3), 263 (10.6).

2.8. *n*-Hexadecanyl Oleate (5)

Elution of the column with petroleum ether – chloroform (1:1) afforded a semisolid mass of **5**, 163 mg, UV λmax (MeOH): 212 nm; IR γ_{max} (KBr): 2925, 2845, 1731, 1643, 1455, 1372, 1248, 1173, 1121, 723 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 5.33 (2H, m, H-9, H-10), 4.19 (2H, t, *J* = 7.2 Hz, H_2-1'), 2.37 (2H, t, *J* = 7.5 Hz, H_2-2'), 2.15 (2H, m, H_2-8), 2.06 (2H, m, H_2-11), 1.63 (4H, m, 2 x CH_2), 1.34 (8H, br s, 4 × CH_2), 1.28 (12H, br s 6 x CH_2), 1.25 (26H, br s 13 x CH_2), 0.89 (3H, t, *J* = 6.6 Hz, Me-18), 0.83 (3H, t, *J* = 6.3 Hz, Me-16′); ^{13}C NMR ($CDCl_3$): δ 171.53 (C-1), 126.17 (C-9), 123.51 (C-10), 63.16 (C-1′), 32.81 (CH_2), 32.08 (CH_2), 29.83 (19 x CH_2), 29.79 (CH_2), 29.56 (CH_2), 29.35 (CH_2), 29.18 (CH_2), 27.43 (CH_2), 25.21 (CH_2), 22.69 (CH_2), 14.22 (Me-18), 14.08 (Me-16′); ESI MS *m/z* (rel. int.): 506 $[M]^+$ ($C_{34}H_{66}O_2$) (2.7), 281 (9.8), 265 (17.3).

2.9. Stigmasteryl Laurate (6)

Elution of the column with petroleum ether-chloroform (1:3) mixture offered colorless crystals of **6**, recrystallized from chloroform-methanol (1:1), 268 mg; R_f: 0.73 (petroleum ether - chloroform - methanol, 6:3.5:05); m. p., 90 – 91 C; UV λ_{max} (MeOH): 211 nm (log ε 5.8); IR γ_{max} (KBr): 2920, 2852, 1743, 1641, 1463, 1373, 1225, 1173, 801, 734 cm $^{-1}$; ^{1}H NMR ($CDCl_3$): δ 5.35 (1H, m, H-6), 5.13 (1H, m, H-22), 5.01 (1H, m, H-23), 3.51 (1H, brm, $w_{1/2}$ = 16.5 Hz, H-3α), 2.25 (2H, t, *J* = 5.2 Hz, H_2-2′), 1.04 (3H, brs, Me-19), 0.93 (3H, d, *J* = 6.3 Hz, Me-21), 0.87 (3H, d, *J* = 6.6 Hz, Me-26), 0.85 (3H, d, *J* = 6.0 Hz, Me-27), 0.83 (3H, t, *J* = 6.1 Hz, Me-12′), 0.80 (3H, d, *J* = 6.6 Hz, Me-29), 0.67 (3H, brs, Me-18), 2.39 –1.17 (41 H, m, 17 x CH_2, 7 x CH); ^{13}C NMR ($CDCl_3$): δ 36.68 (C-1), 31.08 (C-2), 71.83 (C-3), 41.90 (C-4), 141.07 (C-5), 121.72 (C-6), 31.13 (C-7), 31.08 (C-8), 49.35 (C-9), 38.02 (C-10), 21.07 (C-11), 39.76 (C-12), 41.88 (C-13), 55.96 (C-14), 24.17 (C-15), 28.67 (C-16), 55.36 (C-17), 11.29 (C-18), 19.20 (C-19), 36.68 (C-20), 18.27 (C-21), 138.31 (C-22), 130.81 (C-23), 45.10 (C-24), 27.28 (C-25), 20.31 (C-26), 18.67 (C-27), 23.11 (C-28), 11.25 (C-29), 173.12 (C-1′), 40.04 (C-2′), 31.42 (CH_2), 30.24 (CH_2), 28.52 (2 x CH_2), 28.37 (4 x CH_2), 25.18 (CH_2), 22.63 (CH_2), 13.67 (C-12′); ESI MS *m/z* (rel. int.): 594 $[M]^+$ ($C_{41}H_{70}O_2$) (6.2), 578 (19.3), 411 (37.8), 395 (100), 394 (41.3), 381 (15.2), 271 (10.2), 255 (23.6), 240 (13.2), 213 (26.5), 198 (21.3), 183 (16.2).

2.10. Stigmasteryl Myristate (7)

Further elution of the column with petroleum ether - chloroform (1: 3) mixture gave colorless crystals of **7**, recrystallized form chloroform - methanol (1:1), 151 mg (0.15% yield), R_f: 0.63 (petroleum ether: chloroform: 7:3), m. p. 75 - 76 C, UV λ_{max} (MeOH): 209, 229 nm (log ε 5.2, 3.1); IR γ_{max}(KBr): 2927, 2857, 1737, 1640, 1464, 1375, 1244, 1181, 1106, 1049, 960, 801, 727 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 5.35 (1H, m, H-6), 5.16 (1H, m, H-22), 5.03 (1H, m, H-23), 3.50 (1H, brm, $w_{1/2}$ = 16.5 Hz, H-3α), 2.28 (2H, t, *J* = 7.2 Hz, H-2′), 1.04 (3H, brs, Me-19), 0.93 (3H, d, *J* = 6.2 Hz, Me-21), 0.87 (3H, d, *J* = 6.4 Hz, Me-26), 0.85 (3H, d, *J* = 6.1 Hz, Me-27), 0.83 (3H, t, *J* = 6.3 Hz, Me-14′), 0.80 (3H, d, *J* = 5.6 Hz, Me-29), 2.35 -1.25 (47 H, m, 20 x CH_2, 7 x CH), 0.67 (3H, brs, Me-18); ^{13}C NMR ($CDCl_3$): δ 36.72 (C-1), 31.10 (C-2), 71.81 (C-3), 41.95 (C-4), 140.71 (C-5), 121.69 (C-6), 31.23 (C-7), 31.05 (C-8), 49.53 (C-9), 38.95 (C-10), 23.56 (C-11), 39.19 (C-12), 41.87 (C-13), 55.99 (C-14), 24.09 (C-15), 28.87 (C-16), 55.35 (C-17), 11.37 (C-18), 19.25 (C-19), 36.72 (C-20), 18.37 (C-21), 139.83 (C-22), 125.62 (C-23), 45.08 (C-24), 27.83 (C-25), 20.36 (C-26), 18.87 (C-27), 22.51 (C-28), 11.58 (C-29), 173.12 (C-1′), 40.04 (C-2′), 31.44 (CH_2), 30.18 (CH_2), 28.59 (7 x CH_2), 27.11 (CH_2), 22.68 (CH_2), 14.26 (C-14′); ESI MS *m/z* (rel.int.): 622 $[M]^+$ ($C_{43}H_{74}O_2$) (5.3), 607 (8.3), 411 (37.2), 394 (12.6), 271 (11.3), 255 (21.6), (6.2), 228 (13.2), 213 (38.1) 211 (15.6), 198 (43.2), 173 (65.8), 159 (90.3), 145 (87.6), 133 (73.2), 192 (7.1), 178 (9.2), 164 (12.5), 122 (3.1), 108 (12.6), 95 (100).

2.11. *n*-Tetracontan-7-One (8)

Elution of the column with chloroform produced colorless crystals of **8**, 134 mg, m. p. 89 – 91 °C, UV λmax (MeOH): 212 nm; IR γ_{max} (KBr): 2925, 2841, 1709, 1637, 1461, 1376, 1255, 1172, 1066, 725 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 2.33 (2H, m, H_2-6), 2.12 (2 H, m, H_2-8), 1.63 (2 H, m, CH_2), 1.55 (2 H, m, CH_2), 1.32 (8 H, m, 4 x CH_2), 1.29 (14 H, br s, 7 x CH_2), 1.24 (44 H, br s, 22 x CH_2), 0.87 (3 H, t, *J* = 6.3 Hz, Me-1), 0.82 (3 H, t, *J* = 6.6 Hz, Me-40); ^{13}C NMR ($CDCl_3$): δ 195.73 (C-7), 31.92 (CH_2), 31.81 (CH_2), 29.70 (33 x CH_2), 29.35 (CH_2), 22.67 (CH_2), 13.22 (Me-1), 12.91 (Me-40); ESI MS *m/z* (rel. int.): 576 $[M]^+$ ($C_{40}H_{80}O$) (5.8), 491 (11.3), 463 (6.2).

2.12. *n*-Pentacos-13′-enyl Oleate (9)

Further elution of the column with chloroform offered light yellow crystals of **9**, recrystallized from acetone - methanol (1:1), 210 mg (0.21% yield); R_f: 0.63 (petroleum ether –chloroform - methanol, 5:4:1); m. p. 63-64 C; IR γ_{max} (KBr): 2917, 2849, 1733, 1644, 1463, 1265, 1097, 912, 804, 721 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 5.67 (1H, m, H-13′), 5.37 (1H, m, H-14′), 5.21 (1H, m, H-9), 5.16 (1H, m, H-10), 3.98 (2H, t, *J* = 6.8 Hz, H_2-1′), 2.24 (2H, t, J = 7.2 Hz, H_2-2), 2.06 (2H, m, H_2-11) 1.98 (2H, m, H_2-8), 1.96 (4H, m, H_2-12′, H_2-15′), 1.53 (4H, m, 2 x CH_2), 1.18 (56H, brs, 28 x CH_2), 0.83 (3H, t, *J* = 6.3 Hz, Me-18), 0.78 (3H, t, *J* = 6.1 Hz, Me-25′). ^{13}C NMR ($CDCl_3$): δ 173.16 (C-1), 136.06 (C-9), 125.87 (C-10), 120.08 (C-13′), 114.06 (C -14′), 62.81 (C -1′), 51.25 (CH_2), 33.82 (CH_2), 31.93 (CH_2), 29.70 (31 x CH_2), 22.69 (CH_2), 14.11 (Me -18), 14.09 (C-25′); ESI MS *m/z* (rel int): 630 $[M]^+$ ($C_{43}H_{82}O_2$) (1.3), 365 (100), 265 (12.2), 181 (16.5), 155 (25.1).

2.13. β-Sitosterol-3β-O-glucoside (10)

Elution of the column with chloroform: methanol (19: 1) produced colorless amorphous powder of **10**, recrystallized from methanol; 275 mg, R_f: 0.72 (chloroform: methanol, 9.3:0.7); m. p. 275-277 C; UV λ_{max} (MeOH): 241 nm (log ε 2.9); IR γ_{max} (KBr): 3401, 3321, 2918, 2849, 1654, 1377, 1261, 1172, 1082 cm^{-1}; ^1H NMR (DMSO-d_6): δ 5.32 (1H, m, H-6), 5.14 (1H, d, *J* = 7.5 Hz, H-1′), 4.81 (1H, m, H-5′), 3.76 (1H, m, H-2′), 3.54 (1H, m, H-3′), 3.43 (1H, m, H-4′), 3.57 (1H, brs, $w_{1/2}$ = 18.5 Hz, H-3), 3.17 (2H, d, *J* = 8.0 Hz, H_2- 6′), 1.04 (3H, brs, Me-19), 0.95 (3H, d, *J* = 6.5 Hz, Me-21), 0.85 (3H, d, *J* = 6.5 Hz, Me-26), 0.82 (3H, d, *J* = 6.3 Hz, Me-27), 0.77 (3H, t, *J* = 7.0 Hz, Me-29), 0.66 (3H, brs, Me-18), 2.65 - 1.05 (29H, m, 11 × CH_2, 7 × CH); ^{13}C NMR ($CDCl_3$): δ 38.63 (C-1), 33.85 (C-2), 73.46 (C-3), 42.27 (C-4), 140.18 (C-5), 122.21 (C-6), 36.18 (C-7), 31.89 (C-8), 50.17 (C-9), 37.19 (C-10), 25.26 (C-11), 39.71 (C-12), 42.27 (C-13), 56.69 (C-14), 24.29 (C-15), 29.34 (C-16), 55.98 (C-17), 11.76 (C-18), 19.74 (C-19), 36.69 (C-20), 21.19 (C-21), 29.14 (C-22), 28.25 (C-23), 45.81 (C-24), 29.67 (C-25), 18.94 (C-26), 19.36 (C-27), 22.94 (C-28), 11.86 (C-29), 101.95 (C-1'), 76.26 (C-2'), 75.63 (C-3'), 69.97 (C-4'), 79.24 (C-5'), 61.78 (C- 6'); ESI MS *m/z* (rel. int.): 576 $[M]^+$ ($C_{35}H_{60}O_6$) (44.4), 413 (31.5), 179 (10.3).

2.14. Lupenyl 3β-O-Arabinpyranosyl 2′-Oleate (11)

Elution of the column with chloroform – methanol (9: 3) furnished colorless crystals of **11**, recrystallized from chloroform – methanol (1: 1), 251 mg, m. p. 221 – 224 °C, UV λmax (MeOH): 213 nm; IR γ_{max} (KBr): 3421, 3328, 2928, 2843, 1732, 1645, 1465, 1378, 1182, 725 cm^{-1}; ^1H NMR ($CDCl_3$): δ 5.35 (1H, d, *J* = 5.6 Hz, H-12), 5.32 (1H, m, H-9"), 5.29 (1H, m, H-10"), 5.17 (1H, d, *J* = 7.5 Hz, H-1′), 4.71 (1H, br s, H_2 - 29a), 4.66 (1H, br s, H_2 –29b), 4.53 (1H, m, H-2′), 4.54 (1H, dd, *J* = 5.4, 7.5 Hz, H-2′), 4.09 (1 H, m, H-3′), 4.03 (1 H, dd, *J* = 5.3, 8.7 Hz, H-3α), 3.84 (2H, d, *J* = 7.8 Hz, H_2-5′), 3.46 (1H, m, H-4′), 2.33 (2H, t, *J* = 7.2 Hz, H_2-2"), 2.25 (2 H, m, H_2-8), 2.15 (2H, m, H_2-11), 1.66 (3H, br s, Me-30), 1.03 (3H, br s, Me-23), 0.97 (3H, br s, Me-25), 0.88 (3H, br s, Me-27), 0.86 (3H, br s, Me-28), 0.83 (3H, br s, Me-26), 0.81 (3H, br s, Me-24), 0.78 (3H, t, *J* = 6.3 Hz, Me-18"); ^{13}C-NMR ($CDCl_3$): δ 36.15 (C-1), 26.51 (C-2), 79.06 (C-3), 36.51 (C-4), 48.77 (C-5), 18.23 (C-6), 32.06 (C-7), 39.45 (C-8), 47.95 (C-9), 37.32 (C-10), 22.68 (C-11), 124.11 (C-12), 139.76 (C-13), 45.31 (C-14), 28.61 (C-15), 35.08 (C-16), 56.73 (C-17), 52.31 (C-18), 45.35 (C-19), 156.99 (C-20), 31.37 (C-21), 42.17 (C-22), 29.26 (C-23), 15.27 (C-24), 18.46 (C-25), 21.11 (C-26), 25.24 (C-27), 21.05 (C-28), 109.15 (C-29), 19.34 (C-30), 105.89 (C-1′), 80.33 (C-2′), 64.43 (C-3′), 68.31 (C-4′), 60.26 (C-5′), 173.85 (C-1″), 34.82 (C-2″), 29.78 (C-3″), 29.76 (C-4″), 29.69 (C-5″), 29.61 (C-6″), 29.58 (C-7″), 34.46 (C-8″), 130.09 (C-9″), 129.82 (C-10″), 32.81 (C-11″), 29.43 (C-12″), 29.32 (C-13″), 29.22 (C-14″), 28.65 (C-15″), 27.70 (C-16″), 22.67 (C-17″), 14.18 (C-18″); ESI MS *m/z* (rel. int.): 820 $[M]^+$ ($C_{53}H_{88}O_6$) (2.3), 423 (21.2), 281 (35.2), 265 (8.3), 216 (11.2), 207 (18.5), 132 (6.1) .

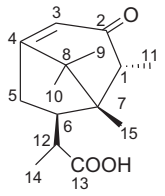
3. RESULTS AND DISCUSSION

The compounds **3**, **4** and **5** were the common fatty esters characterized as *n*-pentadecanyl octadec-9, 12- dienoate (*n*-pentadecanyl linoleate, **3)**, *n*-hexadecanyl linoleate (**4**) and *n*-hexadecanyl oleate (**5**) (Fig. **1**).

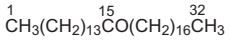
Compound **1** yielded effervescences with sodium bicarbonate solution and exhibited IR absorption bands for carbonyl group (1703 cm^{-1}), unsaturation (1645 cm^{-1}) and carboxylic function (3363, 1685 cm^{-1}). On the basis of mass and ^{13}C NMR spectra, the molecular ion peak of **1** was established at *m/z* 250 consistent with a molecular formula of a sesquiterpene, $C_{15}H_{22}O_3$. The ^1H NMR spectrum of **1** showed a one-proton singlet at δ 5.72 assigned to vinylic H-3 proton. Three singlets at δ 1.25, 1.21 and 0.83 and two doublets at δ 1.14 (*J* = 6.2 Hz) and 0.65 (*J* = 6.5 Hz), all integrated for three protons each, were attributed to tertiary C-9, C-10 and C-15 and secondary C-14 and C-11 methyl protons located on saturated carbons. The remaining methine and methylene protons resonated from δ 2.59 to 1.62. The ^{13}C NMR spectrum of **1** exhibited signals for the carboxylic carbon at δ 181.93 (C-13), carbonyl carbon at δ 210.98 (C-2), vinylic carbons at δ 145.66 (C-2) and 133.16 (C-3) and methyl carbons at δ 24.83 (C-9), 26.98 (C-10), 19.57 (C-11), 16.88 (C-14) and 8.41 (C-15). On the basis of these evidences the structure of **1** has been established as 12-methyl cyprot-3-en-2-one-13-oic acid, a new cyprotene-type sesquiterpene (Fig. **1**).

Compound **2** showed its IR absorption bands for carbonyl group (1708 cm^{-1}) and long aliphatic chain (723 cm^{-1}). Its mass spectrum displayed a molecular ion peak at *m/z* 464 corresponding to a molecular formula of an aliphatic ketone, $C_{32}H_{64}O$. The ion peaks generating at *m/z* 225 [C_{15} – C_{16} fission, CH_3 $(CH_2)_{13}CO]^+$ and 177 [C_{14} – C_{15} fission, CH_3 $(CH_2)_{13}]^+$ suggested the presence of the carbonyl function at C_{15} carbon. The ^1H NMR spectrum of **2** exhibited five two-proton multiplets from δ 2.34 to 1.33 and two broad singlets at δ 1.28 (6H) and 1.23 (42 H) assigned to methylene

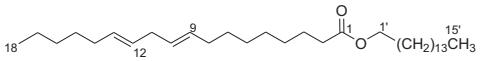
protons. Two three-proton triplets at δ 0.86 (J = 6.6 Hz), 0.83 (J = 6.1 Hz) were accounted to terminal C-32 and C-1 primary methyl protons, respectively. The ^{13}C NMR spectrum of **2** displayed signals for the carbonyl carbon at δ 191.75 (C-15), methylene carbons between δ 31.98 - 22.79 and methyl carbons at δ 14.89 (C-1) and 14.09 (C-32). The absence of any signal beyond δ 2.34 in the ^{1}H NMR spectrum and between δ 191.75 - 32.16 in the ^{13}C NMR spectrum ruled out the unsaturated nature of the molecule. On the basis of foregoing spectral data analysis, the structure of **2** has been elucidated as *n*-dotriacontan-15-one, a new aliphatic ketone (Fig. **1**).



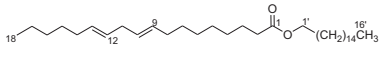
12-Methyl cyprot-3-en-2-one-13-oic acid **(1)**



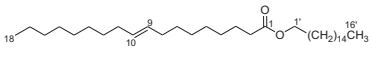
n-Dotriacontan-15-one **(2)**



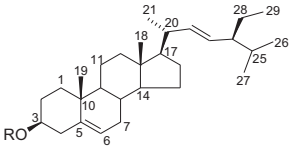
n-Pentadecanyl linoleate **(3)**



n-Hexadecanyl linoleate **(4)**

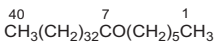


n-Hexadecanyl oleate **(5)**

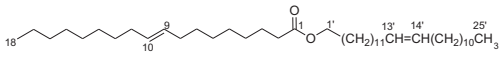


R = $CO(CH_2)_{10}CH_3$ Stigmasterol *n*-dodecanoate **(6)**

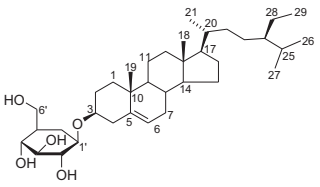
R = $CO(CH_2)_{12}CH_3$ Stigmasterol *n*-tetradecanoate **(7)**



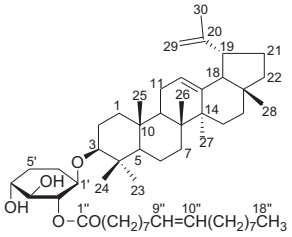
n-Tetracontan-7-one **(8)**



n-Pentacos-13′-enyl oleate **(9)**



β-Sitosterol glucoside **(10)**



Lupenyl arabinopyranosyl oleate **(11)**

Fig. (1). Structural formulae of the chemical constituents **1** – **11**.

Compound **6** gave positive tests of steroids and demonstrated IR absorption bands for ester group (1743 cm^{-1}), unsaturation (1641 cm^{-1}) and long aliphatic chain (734 cm^{-1}). On the basis of mass and ^{13}C NMR spectra, its molecular weight was established at *m/z* 594 consistent with the molecular formula of a steroid esterified with C_{12} fatty acid, $C_{41}H_{70}O_2$. The prominent ion peaks generated at *m/z* 578 $[M - Me]^+$, 183 $[CO(CH_2)_{10}CH_3]^+$, 411 $[M - 183]^+$, 395 $[411\text{-}Me]^+$, 394 $[411\text{-}OH]^+$, 271 $[411 - C_{10}H_{19}$, side chain$]^+$, 255 $[271 - Me]^+$, 240 $[255 - Me]^+$, 213 $[255$ - ring D fission$]^+$ and 198 $[213 - Me]^+$ suggested that a C_{12} acyl moiety was esterified with a sterol containing a C_{10} unsaturated side chain. The ^{1}H NMR spectrum of **6** displayed three one-proton multiplets at δ 5.35, 5.13 and 5.01 assigned to vinylic H-6, H-22 and H-23 protons, respectively. A one-proton broad multiplet at δ 3.51 with half width of 16.5 Hz was attributed to α-oriented oxymethine H-3 proton. Two broad singlets at δ 0.67 and 1.04, four doublets at δ 0.93 (J = 6.3 Hz), 0.87 (J = 6.6 Hz), 0.85 (J = 6.0 Hz) and 0.80 (J = 6.6 Hz) and a triplet at δ 0.83 (J = 6.1 Hz), all integrating for three protons each, were accounted to tertiary C-18 and C-19, secondary C-21, C-26 and C-27 and primary C-29 and C-12′ methyl

protons, respectively, all attached to the saturated carbons. A two-proton triplet at δ 2.25 (J = 5.2 Hz) was due to C-2′ methylene protons adjacent to the ester group. The remaining methylene and methine protons resonated between δ 2.39 - 1.17. The ^{13}C NMR spectrum of **6** showed important signals for the ester carbon at δ 173.12 (C-1′), vinylic carbons at δ 141.07 (C-5), 121.72 (C-6), 138.31 (C-22) and 130.81 (C-23), oxymethine carbon at δ 71.83 (C-3) and the other methyl, methylene and methine carbons between δ 55.36 - 11.25. Alkaline hydrolysis of **6** yielded stigmasterol, m. p. 162 - 164 °C, and lauric acid, m. p. 43 °C, R_f 0.24 (glacial acetic acid, 85%). The ^{1}H NMR and ^{13}C NMR spectral data of the steroidal nucleus were compared with other stigmastene-type molecules [35, 36]. On the basis of spectral data analysis and chemical reactions, the structure of **6** has been established stigmast-5,22–dien-3β –olyl *n*-dodecanoate (stigmasterol laurate), a rare sterol ester present in tobacco smoke [37] (Fig. **1**).

Compound **7** had IR absorption bands for ester group (1737 cm^{-1}), unsaturation (1640 cm^{-1}) and long aliphatic chain (727 cm^{-1}). On the basis of mass and ^{13}C NMR spectra, its molecular weight was determined at *m/z* 622 consistent with the molecular formula of a sterol ester $C_{43}H_{74}O_2$. The prominent ion fragments generating at *m/z* 607 [M - Me]$^+$, 211 $[CH_3(CH_2)_{12}CO]^+$, 228 $[CH_3\ (CH_2)_{12}\ COOH]^+$, 411 $[M - CO(CH_2)_{12}CH_3]^+$, 394 $[M - CH_3(CH_2)_{12}COOH]^+$, 271 [411-$C_{10}H_{19}$, side chain]$^+$, 255 [271 - Me]$^+$, 240 [255 - Me]$^+$, 213 [255 - ring D fission]$^+$ and 198 [213 – Me]$^+$ suggested that the sterol was esterified with a C_{14} fatty acid. The ion peaks produced at *m/z* 108 [$C_{6,7}$ - $C_{9,10}$ fission]$_+$, 122 [$C_{7,8}$ - $C_{9,10}$ fission]$^+$, 164 [$C_{8,14}$ - $C_{9,11}$ fission]$^+$, 178 [$C_{8,14}$ - $C_{11,12}$ fission]$^+$ and 192 [$C_{8,14}$ - $C_{12,13}$ fission]$^+$ supported the existence of one of the vinylic linkage in ring B at C_5 and saturated nature of the ring C. The ^{1}H NMR spectrum of **7** displayed three one - proton multiplets at δ 5.35, 5.16 and 5.03 ascribed to vinylic H-6, H-22 and H-23 protons, respectively. A one-proton broad multiplet at δ 3.50 with half-width of 16.5 Hz was attributed to *α*–oriented oxymethine H-3 proton. The methyl protons appeared as three – proton singlets at δ 1.04 (Me-19) and 0.67 (Me-18), as doublets at δ 0.93 (J = 6.2 Hz, Me-21), 0.87 (J = 6.4 Hz, Me-26), 0.85 (J = 6.1 Hz, Me-27) and 0.80 (J = 5.6 Hz, Me-29) and as a triplet at δ 0.83 (J = 6.3 Hz, Me-14′), The remaining methine and methylene protons resonated in the range δ 2.35 - 1.17. The ^{13}C NMR spectrum of **7** showed important signals for ester carbon at δ 173.12 (C-1′), vinylic carbons at *δ* 140.71 (C-5), 121.69 (C-6), 139.83 (C-22) and 125.62 (C-23), oxymethine carbon at δ 71.81 (C-3) and methyl carbons between δ 22.50 – 11.37. The ^{1}H NMR and ^{13}C NMR spectral data of the steroidal nucleus of **7** were compared with reported data of related steroids [35, 36]. Alkaline hydrolysis of **7** afforded stigmasterol, m. p. 162 - 164 °C and myristic acid, m. p. 54 °C, R_f 0.58 (*n*-hexane). On the basis of the foregoing discussion the structure of **7** has been elucidated as stigmast-5, 22-dien-3*β*-olyl *n*-tetradecanoate (stigmasterol myristate), a unique sterol ester present in tobacco smoke [37] (Fig. **1**).

Compound **8** showed IR absorption bands for carbonyl group (1709 cm^{-1}) and long aliphatic chain (725 cm^{-1}). Its mass spectrum had a molecular ion peak at *m/z* 576 corresponding to a molecular formula of an aliphatic ketone, $C_{40}H_{80}O$. The ion peaks produced at *m/z* 491 [$C_7 – C_6$ fission, $CH_3\ (CH_2)_5CO$] $^+$ and 463 [$C_7 – C_8$ fission, $CH_3\ (CH_2)_{32}$] $^+$ suggested the presence of the carbonyl function at C_7 carbon. The ^{1}H NMR spectrum of **8** exhibited three two-proton multiplets at δ 2.33, 2.09 and 1.55 and four broad singlets at δ 1.63 (4H), 1.32 (8H), 1.29 (12H) and 1.23 (44 H) assigned to methylene protons. Two three-proton triplets at δ 0.87 (J = 6.3 Hz) and 0.82 (J = 6.6 Hz) were accounted to terminal C-1 and C-40 primary methyl protons, respectively. The ^{13}C NMR spectrum of **8** displayed signals for the carbonyl carbon at δ 195.73 (C - 7), methylene carbons between δ 31.92 - 22.67 and methyl carbons at δ 13.22 (C-1) and 12.91 (C-40). The absence of any signal beyond δ 2.33 in the ^{1}H NMR spectrum and between δ 195.73 - 31.92 in the ^{13}C NMR spectrum supported saturated nature of the molecule. On the basis of foregoing spectral data analysis, the structure of **8** has been elucidated as *n*-tetracontan-7-one, a new aliphatic ketone (Fig. **1**).

Compound **9** showed IR absorption bands for ester group (1733 cm^{-1}), unsaturation (1644 cm^{-1}) and long aliphatic chain (721 cm^{-1}). On the basis of mass spectrum the molecular ion peak of **9** was determined at *m/z* 630 consistent with the molecular formula of a fatty acid ester, $C_{43}H_{82}O_2$. The generation of the ion peaks at *m/z* 365 $[CH_3\ (CH_2)_{10}CH{=}CH\text{-}(CH_2)_{12}O]^+$ and 265 $[CH_3(CH_2)_7CH{=}CH(CH_2)_7CO]^+$ indicated that a C_{25} alcohol was esterified with a C_{18}-fatty acid. The formation of the ion peaks at *m/z* 155 $[CH_3\ (CH_2)_{10}]^+$ and 181 $[CH_3(CH_2)_{10}CH{=}CH]^+$ supported the location of one of the vinylic linkage at C-14′. The ^{1}H NMR spectrum of **9** showed four one–proton multiplets at δ 5.67, 5.37, 5.21 and 5.16 assigned to vinylic H-13′, H-14′, H-9 and H-10 protons, respectively. Two two-proton triplets at δ 3.98 (J = 6.8 Hz) and 2.24 (J = 7.2 Hz) were ascribed to oxymethylene H_2-1′ protons and methylene H_2-2 protons adjacent to the ester group, respectively. The remaining methylene protons appeared between *δ* 2.06 - 1.18. Two three–proton triplets at δ 0.83 (J = 6.3 Hz) and 0.78 (J = 6.1 Hz) were due to C-18 and C-25′ primary methyl protons, respectively. The ^{13}C NMR spectrum of **9** showed signals for ester carbon at δ 173.16 (C-1), vinylic carbons at δ 136.06 (C-9), 125.87 (C-10), 120.08 (C-13′) and 114.06 (C -14′), oxymethylene carbon at 62.81 (C-1′), other methylene carbons between δ 51.25 - 22.69 and methyl

carbons at δ 14.11 (C-18) and 14.09 (C-25′). On the basis of the foregoing account, the structure of **9** has been formulated as *n*-pentacos-13′-enyl *n*-octadec-9-enoate (*n*-pentacos-13′-enyl oleate), a new fatty ester (Fig. **1**).

The compound **10** was a known steroidal glycoside characterized as β-sitosterol-3β-O-glucoside [38, 39].

Compound **11** gave positive tests for triterpenic glycoside and showed characteristic IR absorption bands for hydroxyl groups (3421, 3328 cm^{-1}), ester function (1732 cm^{-1}), unsaturation (1645 cm^{-1}) and long aliphatic chain (725 cm^{-1}). Its molecular ion peak was determined on the basis of mass and ^{13}C NMR spectra at *m/z* 820 consistent to a molecular formula of a triterpenic glycosidic ester, $C_{53}H_{88}O_6$. The ion peaks arising at *m/z* 265 [$C_{1''}$ – O fission, $CH_3(CH_2)_7$-CH=CH-$(CH_2)_7CO]^+$, 281 [$C_{1'}$ – O fission, $CH_3(CH_2)_7$-CH=CH-$(CH_2)_7$ $COO]^+$ and 423 [M - $CH_3(CH_2)_7$-CH=CH-$(CH_2)_7CO$-$C_5H_8O_4]^+$ indicated that oleic acid was linked to a pentose unit which are attached to a lupene-type triterpene. The ion peaks produced at *m/z* 207 and 216 due to Retro-Diels Alder fragmentation of ring C of the triterpenic unit suggested that one of the vinylic linkage was present at C_{12} carbon. The ^1H NMR spectrum of **11** exhibited two one - proton doublets at δ 5.35 (*J* = 5.6 Hz) and 5.17 (*J* = 7.5 Hz) and two one - proton multiplets at δ 5.32 and 5.29 assigned to vinylic H-12, anomeric H-1' and vinylic H-9" and H-10" protons, respectively. Two one - proton singlets at δ 4.71 and 4.66 were due to vinylic H_2-29 methylene protons of the lupene-type triterpenic unit. A one - proton double doublet at δ 4.03 (*J* = 5.3, 8.7 Hz) was ascribed to oxymethine H-3α proton. Another one – proton double doublet at δ 4.54 (*J* = 5.4, 7.5 Hz) was accounted to oxymethine H-2′ proton and its deshielding nature suggested the location of ester formation at C-2'. The other sugar proton appeared as one-proton multiplets at δ 4.09 (H-4') and 3.46 (H-4') and as a two - proton doublet at δ 3.84 (*J* = 7.0 Hz, H_2-5'). A two-proton triplet at δ 2.33 (*J* = 7.2 Hz) was due to methylene H_2-2" protons adjacent to the ester group. A three - proton singlet in the deshielded region at δ 1.66 was attributed to C-30 methyl protons located on C-20 vinylic carbon. The other methyl signals appeared as three - proton singlets from δ 1.02 to 0.83. A three - proton triplet at δ 0.78 (*J* = 6.3Hz) was assigned to C-18" primary methyl protons. The ^{13}C NMR spectrum of **11** displayed signals for ester carbon at δ 173.85 (C-1"), vinylic carbons at δ 124.11 (C-12), 139.76 (C-13), 156.99 (C-20), 109.15 (C-29), 130.09 (C-9′′) and 129.82 (C-10′′), anomeric carbon at δ 105.89 (C-1′), oxymethine carbon at δ 79.06 (C-3), and other sugar carbons at δ 80.33 (C-2'), 64.43 (C-3'), 68.31 (C-4'), 60.26 (C-5'). The carbon signals of the triterpenic unit were compared with related lupene-type molecules [40]. Acid hydrolysis of **11** yielded lup-12, 20 (29)-dien-3β-ol, α-L-arabinose and oleic acid, co-TLC comparable. On the basis of spectral data analysis and chemical reactions, the structure of **11** had been formulated as lup-12, 20 (29)-dien-3β-ol-3-α-L-arabinopyranosyl-2'-oleate (lupenyl 3β-O-arabinpyranosyl 2′-oleate). This is a new lupene-type triterpenic glycosidic ester (Fig. **1**).

CONCLUSION

Phytochemical investigation of a methanolic extract of the tubers of *Cyperus rotundus* resulted in the isolation of cyprot-3-en-2-one-14-oic acid, aliphatic ketones, fatty esters, steroidal esters, β-sitosterol-3β-O-glucoside and lupenyl 3β-O-arabinpyranosyl oleate. This work has enhanced understanding about the phytoconstituents of the plant. These compounds may be used as chromatographic markers for standardization of the tubers of the plant.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

HUMAN AND ANIMAL RIGHTS

No Animals/Humans were used for studies that are base of this research.

CONSENT FOR PUBLICATION

Not applicable.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

ACKNOWLEDGEMENTS

The authors are thankful to the instrumentation centers, Central Drug Research Institute, Lucknow and Jawaharlal Nehru University, New Delhi for recording spectral data of the compounds.

REFERENCES

[1] Gunasekera TG, Fernando DN. Agricultural importance, biology, control and utilisation *Cyperus rotundus.* Planter, Kuala Lumpur 1994; 70: 537-44.

[2] Anonymous . The Wealth of India, raw materials, NISCAIR. CSIR, New Delhi 2001; 3: 333-4.

[3] Joshi AR, Joshi K. Indigenous knowledge and uses of medicinal plants by local communities of the Kali Gandaki Watershed Area, Nepal. J Ethnopharmacol 2000; 73(1-2): 175-83.
[http://dx.doi.org/10.1016/S0378-8741(00)00301-9] [PMID: 11025154]

[4] Oliver-Bever B. Medicinal Plants in Tropical West Africa. Cambridge, UK: Cambridge University Press 1986; p. 200.
[http://dx.doi.org/10.1017/CBO9780511753114]

[5] El-Kamali HH, El-Khalifa KF. Folk medicinal plants of riverside forests of the Southern Blue Nile district, Sudan. Fitoterapia 1999; 70: 493-7.
[http://dx.doi.org/10.1016/S0367-326X(99)00073-8]

[6] Singh N, Pandey BR, Verma P, Bhalla M, Gilca M. Phyto-pharmacotherapeutics of *Cyperus rotundus* Linn. (Motha): an overview. Indian J Nat Prod Resour 2012; 3(4): 467-76.

[7] Sivapalan SR. Medicinal uses and parmacological activities of *Cyperus rotundus* Linn. – a review. Int J Sci Res Publications 2013; 3(5): 1-8.

[8] Kirtikar KR, Basu BD. Indian Medicinal Plants. 2 nd ed. International Book Distributors, Dehradun 2007; 4: 878.

[9] Boulos L, El-Hadidi MN. The weed flora of Egypt. Cairo: The American University in Cairo Press 1984; p. 58.

[10] Elizabeth MW. Major Herbs of Ayuveda. London: Elsevier Health Science 2002.

[11] Sonwa MM, König WA. Chemical study of the essential oil of Cyperus rotundus. Phytochemistry 2001; 58(5): 799-810.
[http://dx.doi.org/10.1016/S0031-9422(01)00301-6] [PMID: 11672746]

[12] Aghassi A, Naeemy A, Feizbakhsh A. Chemical composition of the essential oil of *Cyperus rotundus* L. from Iran. J Essen Oil Bearing Plants 2013; 16(3): 382-6.
[http://dx.doi.org/10.1080/0972060X.2013.794041]

[13] Ekundayo O, Oderinde R, Ogundeyin M, Stahl-Biskup E. Essential oil constituents of *Cyperus tuberosus* Rottb. rhizomes. Fla Frag J 1991; 6(4): 261-4.
[http://dx.doi.org/10.1002/ffj.2730060404]

[14] Lawal OA, Oyedeji AO. Chemical composition of the essential oils of *Cyperus rotundus* L. from South Africa. Molecules 2009; 14(8): 2909-17.
[http://dx.doi.org/10.3390/molecules14082909] [PMID: 19701133]

[15] Kilani S, Ledauphin J, Bouhlel I, *et al.* Comparative study of *Cyperus rotundus* essential oil by a modified GC/MS analysis method. Evaluation of its antioxidant, cytotoxic, and apoptotic effects. Chem Biodivers 2008; 5(5): 729-42.
[http://dx.doi.org/10.1002/cbdv.200890069] [PMID: 18493959]

[16] Kim SJ, Ryu B, Kim HY, *et al.* Sesquiterpenes from the rhizomes of *Cyperus rotundus* and their potential to inhibit LPS-induced nitric oxide production. Bull Korean Chem Soc 2013; 34: 2207-10.
[http://dx.doi.org/10.5012/bkcs.2013.34.7.2207]

[17] Bisht A, Bisht GR, Singh M, Gupta R, Singh V. Chemical composition and antimicrobial activity of essential oil of tubers of *Cyperus rotundus* Linn. collected from Dehradun (Uttarakhand). Int J Res Pharm Biomed Sci 2011; 2: 661-5.

[18] Tiwari R, Kumar S. Chemical constituents of the essential oil of *Cyperus rotundus* Linn. Int J Drug Develop Res 2014; 6(2): 57-60.

[19] Nidugala H, Avadhani R, Prabhu A, Basavaiah R, Sunil Kumar KN. GC-MS characterization of n-hexane soluble compounds of *Cyperus rotundus* L. rhizomes. J Appl Pharm Sci 2015; 5(12): 96-100.
[http://dx.doi.org/10.7324/JAPS.2015.501216]

[20] Yoshizaki Y, Nakagawa J, Ando T, Hisamichi S. Simplified determination of sugetriol triacetate in cyperi rhizoma. Shoyakugaku Zasshi 1992; 46: 187-9.

[21] Kalsi P, Sharma A, Singh A, Singh IP, Chhabra BR. Biogenetically important sesquiterpenes from *Cyperus rotundus*. Fitoterapia 1995; 66: 94.

[22] Thebtaranonth C, Thebtaranonth Y, Wanauppathamkul S, Yuthavong Y. Antimalarial sesquiterpenes from tubers of *Cyperus rotundus*: structure of 10,12-peroxycalamenene, a sesquiterpene endoperoxide. Phytochemistry 1995; 40(1): 125-8.
[http://dx.doi.org/10.1016/0031-9422(95)00260-E] [PMID: 7546546]

[23] Ohira S, Hasegawa T, Hayashi KI, Hoshino T, Takaoka D, Nozaki H. Sesquiterpenoids from *Cyperus rotundus*. Phytochemistry 1998; 47: 1577-81.
[http://dx.doi.org/10.1016/S0031-9422(97)00825-X]

[24] Singh PN, Singh SB. A new saponin from mature tubers of *Cyperus rotundus.* Phytochemistry 1980; 19: 2056.
[http://dx.doi.org/10.1016/0031-9422(80)83046-9]

[25] Jeong SJ, Miyamoto T, Inagaki M, Kim YC, Higuchi R. Rotundines A-C, three novel sesquiterpene alkaloids from *Cyperus rotundus*. J Nat

Prod 2000; 63(5): 673-5.
[http://dx.doi.org/10.1021/np990588r] [PMID: 10843585]

[26] Sayed HM, Mohamed MH, Farag SF, Mohamed GA. Phytochemical and biological investigations of *Cyperus rotundus* L. Bull Fac Pharm Cairo Univ 2001; 39: 195-203.

[27] El-Habashy L, Mansour RM, Zahran MA, El-Hadidi MN, Saleh NA. Leaf flavonoids of Cyperus species in Egypt. Biochem Syst Ecol 1989; 17: 191-5.
[http://dx.doi.org/10.1016/0305-1978(89)90078-1]

[28] Krishna S, Renu S. Isolation and identification of flavonoids from *Cyperus rotundus* Linn. *in vivo* and *in vitro*. J Drug Deliv Ther 2013; 3: 109-13.

[29] Sayed HM, Mohamed MH, Farag SF, Mohamed GA, Omobuwajo OR, Proksch P. Fructose-amino acid conjugate and other constituents from *Cyperus rotundus* L. Nat Prod Res 2008; 22(17): 1487-97.
[http://dx.doi.org/10.1080/14786410802038556] [PMID: 19023813]

[30] Koichiro K, Kunikazu U. Secondary metabolic compounds in purple nutsedge (*Cyperus rotundus L.*) and their plant growth inhibition. Shokubutsu no Kagaku Chosetsu 1981; 16: 32-7.

[31] Zhou Z, Zhang H. Phenolic and iridoid glycosides from the rhizomes of *Cyperus rotundus* L. Med Chem Res 2013; 22: 4830-5.
[http://dx.doi.org/10.1007/s00044-013-0504-9]

[32] Mohamed GA. Iridoids and other constituents from *Cyperus rotundus* L. rhizomes. Bull Fac Pharm Cairo Univ 2015; 53: 5-9.
[http://dx.doi.org/10.1016/j.bfopcu.2015.01.001]

[33] Sayed HM, Mohamed MH, Farag SF, Mohamed GA, Proksch P. A new steroid glycoside and furochromones from *Cyperus rotundus* L. Nat Prod Res 2007; 21(4): 343-50.
[http://dx.doi.org/10.1080/14786410701193056] [PMID: 17479423]

[34] Alam P, Ali M, Aeri V. Isolation of keto alcohol and triterpenes from tubers of *Cyperus rotundus* Linn. J Nat Prod Plant Resour 2012; 2(2): 272-80.

[35] Jung WS, Chung IM, Ali M, Ahmad A. New steroidal glycoside ester and aliphatic acid from the fruits of Lycium chinense. J Asian Nat Prod Res 2012; 14(4): 301-7.
[http://dx.doi.org/10.1080/10286020.2011.653346] [PMID: 22375866]

[36] Mustafa A, Ali M. New steroidal lactones and homomonoterpenic glucoside from fruits of *Malva sylvestris* L. Acta Pol Pharm 2011; 68(3): 393-401.
[PMID: 21648194]

[37] Rodgman A, Perfetti TA. The Chemical Components of Tobacco and Tobacco Smoke. 2nd ed. Boca Raton, Florida: CRC Press 2013; p. 241.
[http://dx.doi.org/10.1201/b13973]

[38] Ali M, Sultana S. Phytochemical investigation of the leaves of *Ficus pandurata* Sander. Eur J Biomed Pharmac Sci 2016; 3(10): 393-6.

[39] Ali A, Jameel M, Ali M. New Naphthyl Esters from the Bark of *Ficus religiosa* Linn. Nat Prod J 2014; 4: 248-53.

[40] Aeri V, Alam P, Ali M, Naquvi KJ. Lupene-type triterpenic and steroidal constituents from the roots of *Streblus asper* Lour. J Sci Innov Res 2015; 4(3): 142-5.