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Chemical Constituents from the Stem barks of Ficus racemosa L. and Ficus religiosa L.

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Abstract *Ficus racemosa* L. and *Ficus religiosa* L. (Moraceae) are distributed in south-eastern Asia, Middle East, Egypt, Libya and the United States. The stem bark of *F. racemosa* is used to treat asthma, burns, diabetes, diarrhoea, dysentery, gynaecological disorders, haematuria, haemoptysis, leucorrhoea, menorrhagia, mouth problems and piles. The stem bark of *F. religiosa* is used to cure blisters, boils, diabetes, diarrhoea, dysentery, fistula, glandular swellings of the neck, gonorrhea, gout, inflammation, jaundice, mouth sores, scabies, ulcers and wounds; to check excess quantity of urine and bleeding, to stabilize fetus and sexual power. The air-dried stem barks were exhaustively extracted with methanol individually in a Soxhlet apparatus. The concentrated methanol extracts were adsorbed on silica gel for column and chromatographed over silica gel column separately. The columns were eluted with petroleum ether, chloroform and methanol successively to isolate the phytoconstituents.

Phytochemical investigation of the stem bark of *F. racemosa* afforded glyceryl 1-oleio-2,3-distearate (glyceryl linolyldistearin, 1), glyceryl 1- linoleiyl-2-oleio-3-phosphate (2), glyceryl 1,2-dioleio-3-phosphate (3), gallic acid (4), (Z)-*n*-hexatriacont-12-en-1-ol (5), lup-12,20(29)-dien-6 β -ol-3 β -olyl oleate (6 β -hydroxylupdienolyl 3 β -oleate, 6), stigmast-5-en-4 α -ol-26-oic acid-3 β -olyl linolenate (stigmast-5-en-6 α -ol-26-oic acid 3 β -linolenate, 7) and β -D-glucuropyranosyl-(6 \rightarrow 1')-O- β -D-glucopyranoside (glucuronosyl-(6 \rightarrow 1')-glucoside, 8). The stem bark of *F. religiosa* furnished arachidic acid (9), glyceryl-1-oleiyl-2-palmityl-3-stearate (10), β -D-arabinopyranosyl 1,2,3-trioctadec-9-enoate (1,2,3-trioleiyl β -D-arabinoside, 11), glyceryl 1-oleio-2-stearyl-3-phosphate (12), glyceryl 1,2-distearyl-3-phosphate (13), glyceryl 1-linoleiyl-2-arachidyl-3-phosphate (14) and 2-(2'-octadec-9''-enoyl β -D-glucopyranosyl) gallic acid [2-(2'-oleiyl glucosyl) gallic acid , (15). The structures of these phytoconstituents have been established on the basis of spectral data analysis and chemical reactions.

Keywords Ficus racemosa, Ficus religiosa, stem barks, phytoconstituents, isolation, characterization.

Introduction

Ficus racemosa L., syn. *Ficus glomerata* Roxb., *Ficus lanceolata* Buch.-Ham. ex Roxb. (Moraceae), known as gular, cluster fig, Indian fig and redwood fig, is a native to India, Australia, Indo-China and New Guinea. It is an evergreen, spreading, lactiferous, deciduous, 15-18 m high tree, without prominent aerial roots; young shoots glabrous, pubescent or scaberulous; leaves are dark green colored, ovate oblong or elliptic-lanceolate, tapering at the apex, margins entire, base acute or rounded, 3-nerved; inflorescence hypanthodium, globose to pyriform, pubescent, peduncled, androgynous solitary or binate, borne on the short, leafless scaly branchlets; flowers staminate, florets



sessile, situated near the mouth of the receptacles; fruits synconus, subglobose or pyriform, orange to red, fleshy [1, 2].

The leaves are used as an astringent and to treat bilious disorders, blisters, boils, bronchitis, chicken pox, diarrhoea, dysmenorrhea, measles, multinodular tuberculosis, ulcers and wounds. A paste of the leaf buds is applied on the skin to improve the complexion [3]. The bark is acrid, astringent, galactagogue, refrigerant, and used to relieve asthma, burns, diabetes, diarrhoea, dysentery, gynaecological disorders, haematuria, haemoptysis, leucorrhoea, menorrhagia, mouth problems and piles [4, 5]. The fruits are astringent, carminative, refrigerant, stomachic, tonic, taken orally to cure asthma, blood disorders, bronchitis, burning sensation, constipation, diabetes, diarrhoea, dry cough, epistaxis, fatigue, haematuria, intestinal worms, diseases of the kidney and spleen, leprosy, leucorrhoea, menorrhagia, stomach pain and haemoptysis [4 - 7]. The fruits ingested with milk promote the growth of fetus and protects from abortion. The unripe fruit is acid, astringent, stypic and tonic, allays thrist, useful in blood diseases, biliousness, coughs and leucorrhoea [4-10]. An exudate from the stem is considered as an aphrodisiac and tonic, applied to cure boils, mumps and other inflammatory glandular enlargements, wounds, and is also used to treat bone fractures, cholera, diarrhoea, diabetes, dysentery, gonorrhea, piles and stomachache. The root is chewed as a treatment for tonsillitis, useful in hydrophobia [4-10]. The fruits contained glauanol, its acetate, hentriacontane, β -sitosterol, glauanol acetate, glucose, gallic, ellagic and tiglic acids, racemosic acid, flavonoids, triterpenoids and tannins [6, 11, 12]. The leaves possessed triterpenoids, glauanol acetate, racemosic acid, tannins, rutin, arabinose, bergapten, psoralenes, flavonoids, ficusin, coumarin, phenolic glycosides and saponins [6, 13-15]. Stem bark afforded gluanol acetate, anthocyanin glycosides, ceryl behenate, α -amyrin acetate, lupeol, friedelin, stigmasterol, β -sitosterol, its Dglucoside, quercetin. bergenin, racemosic acid, α - and β -amyrins and lupeol acetate [6, 16-18].

Ficus religiosa L., syn. F. peepul Griff., Urostigma religiosum (L.) Gasp. (Moraceae),

known as peepal tree, ashwattha tree, sacred fig, bodhi tree, is distributed in India, Bangladesh, China, Myanmar, Pakistan, Thailand, Middle East, Egypt, Libya and the United States. It is a large, deciduous or semi-evergreen tree, up to 30 m tall; trunk 1 to 3 m of diameter; leaves, cordate, ovate, bright, with an extended drip tip; petiole 6–10 cm long; bark flat or slightly curved, grey outside, with thin flakes, inner surface smooth, yellowish to orange, fibrous; fruits small figs, green, ripening to purple [1].

The bark is antidiabetic, astringent and refrigerant, used to check excess quantity of urine and bleeding, to stabilize fetus and sexual power and to cure blisters, boils, diabetes, diarrhoea, dysentery, fistula, glandular swellings of the neck, gonorrhea, gout, inflammation, jaundice, mouth sores, scabies, ulcers and wounds. A milky exudate is applied to calm down body pain and feet cracks. The leaf sap is effective to allay diarrhoea, cholera, gum bleeding and swelling and for wound healing. A leaf paste is lapped to relieve piles; used as an alterative, analgesic, antidote, aphrodisiac, astringent, antigonorrhoeal, purgative and to alleviate asthma, boils, bruises, constipation, coughs, diarrhoea, earache, eye and gastric troubles, fistula, haematuria, haemoptysis, heart diseases, mumps, toothache, migraine, scabies, toothache and wounds [1, 19-22].

The fruit is laxative, alexipharmic and aphrodisiac, used to treat asthma, biliousness, blood diseases, dehydration, dysentery, heart diseases, menstrual disorders, ulcers, uterine troubles and vomiting. The fruit powder with haritaki (*Terminalia chebula*) is taken to control diabetes. The seeds are alterative, laxative and refrigerant; useful in urinary troubles. If the seeds are taken for three days during menstruation, women are sterilized for long time. The roots are utilized to relieve arthritis, rheumatism and gum disease. The aerial roots are diuretic; used to treat ascites and are chewed by women to promote fertility. The root bark is aphrodisiac, astringent; useful to relieve jaundice, leucorrhoea, stomatitis, ulcers and to promote granulations [1, 19-22].

The bark contained phenols, tannins, phytosterols, alkaloids, flavonoids, vitamin K, methyl oleanolate, noctacosanol, lanosterol, lupen-3-one, leucoanthocyanins, bergapten, begaptol, tetratriaconten-2-one, pentatriacontan-5-one, heptatriacontent-10-one and meso –anisosital [23-26]. The roots possessed β -sitosteryl-D-glucoside. The crude latex gave a serine protease religiosin [27]. The stems afforded lupeol, γ -sitosterol and 1,2-benzenediol [28]. The fruits furnished protein, essential amino acids, kaempferol, quercetin, myricetin, undecane,



tridecane, tetradecane, mono- and sesquiterpenes [25, 29-31]. The seeds afforded phytosterolin, β -sitosterol and its glycoside, albuminoids, carbohydrate, fatty matter, coloring matter and caoutchoue. The leaves yielded phytosterols, albuminoids, sugars, fatty matter, caoutchoue, α -amyrin, lupeol, tannic acid, amino acids, higher alkanes, and an essential oil composed mainly of eugenol, itaconic anhydride, 3-methyl-cyclopenetane-1,2-dione, 2-phenylethyl alcohol and benzyl alcohol [23, 25, 32].

Materials and Methods

General Procedures

Melting points were determined on a Perfit melting point apparatus and are uncorrected. UV spectra were measured on Shimadzu-120 double beam spectrophotometer with methanol as a solvent. IR spectra were recorded in KBr pellet on a Shimadzu FTIR-8400 spectrophotometer. The ¹H NMR (300 MHz) and ¹³C NMR (75 MHz) spectra were scanned on a Bruker DRX instruments 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 F254 precoated TLC plates (Merck, Mumbai, India). Spots were visualized by exposing to iodine vapors and UV radiations (254 and 366 nm) and spraying with ceric sulphate solution.

Plant Materials

The stem barks of *Ficus racemosa* and *F. religiosa* were procured from the wild trees located in Ghaziabad (U.P.), India and authenticated by Prof. M. P. Sharma, Taxonomist, Department of Botany, Jamia Hamdard, New Delhi. The voucher specimens of these plant parts are preserved in the herbarium of the Department of Pharmacognosy and Phytochemistry, Jamia Hamdard, New Delhi.

Extraction and Isolation

The plant parts (1 kg each) were coarsely powdered and extracted exhaustively with methanol individually in a Soxhlet apparatus. The extracts were concentrated under reduced pressure to get dark brown masses, 119.6 g and 114.2 g, respectively. The dried residues (100 g each) were dissolved in a minimum amount of methanol and adsorbed on silica gel column grade (60-120 mesh) individually to obtain slurries. Each slurry was air-dried and chromatographed over silica gel columns loaded in petroleum ether (b. p. 60 - 80°C) separately. The columns were eluted with petroleum ether, 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) mixtures. Various fractions were collected singly and matched by TLC to check homogeneity. Similar fractions having the same R_f values were combined and crystallized with solvents. The isolated compounds were recrystallized to get the pure compounds.

Isolation of phytoconstituents from the stem bark of Ficus racemosa

Glyceryl 1- linolyl 2,3-distearin (1)

Elution of the column with petroleum ether yielded a pale yellow semisolid mass of **1**, purified by preparative TLC using petroleum ether - chloroform (1:1), 513 mg, m. p. : 50 - 51 °C; UV λ_{max} (MeOH): 235 nm (log ϵ 2.7); IR υ_{max} (KBr): 2921, 2852, 1737, 1641, 1462, 1378, 1243, 1174, 1095, 1025, 986, 721 cm⁻¹; ¹H NMR (CDCl₃): δ 5.18 (1H, m, H-9'), 5.11 (1H, m, H-10'), 5.09 (2H, m, H-12', H-13'), 4.50 (1H, m, H-2), 4.13 (2H, d, J = 7.2 Hz, H₂-1), 4.05 (2H, d, J = 7.7 Hz, H₂-3), 2.55 (2H, m, H₂-11'), 2.29 (2H, m, H₂-2'), 2.27 (2H, m, H₂-2''), 2.25 (2H, m, H₂-2'''), 2.04 (2H, m, H₂-8'), 1.93 (2H, m, H₂-14'), 1.68 (2H, m, CH₂), 1.62 (2H, m, CH₂), 1.52 (2H, m, CH₂), 1.39 – 1.31 (8H, m, 4 x CH₂), 1.28 (10H, brs, 5 x CH₂), 1.25 (14H, brs, 7 x CH₂), 1.23 (38H, brs, 19 x CH₂), 0.88 (3H, t, J = 6.1 Hz, Me-18''), 0.85 (3H, t, J = 6.1 Hz, Me-18''), 0.82 (3H, t, J = 6.3 Hz, Me-18'''); ¹³C NMR (CDCl₃): δ 173.77 (C-1'), 171.27 (C-1''), 169.68 (C-1'''), 133.58 (C-10'), 130.41 (C-12'), 124.42 (C-9'), 121.74 (C-13'), 80.67 (C-2), 60.22 (C-1), 60.03 (C-3), 55.35 (C-11'), 47.13 (C-2'), 47.04 (C-2''), 46.88 (C-2'''), 42.16 - 39.71 (4 x CH₂), 38.54 – 34.50 (7 x



CH₂), 33.84 - 30.14 (9 x CH₂), 29.80 - 29.28 (4 x CH₂), 28.84 - 28.07 (3 x CH₂), 27.53 - 27.02 (3 x CH₂), 26.85 - 26.05 (4 x CH₂), 25.28 (CH₂), 23.70 - 23.32 (3 x CH₂), 22.79 (3 x CH₂), 15.64 (Me-18'), 14.51 (Me-18''), 14.21 (Me-18'''); EIMS *m*/z (rel. int.): 886 [M]⁺ (C₅₇H₁₀₆O₆) (23.6), 267 (11.6), 263 (9.4).

Glyceryl 1- linolyl-2-oleio-3-phosphate (2)

Elution of the column with chloroform pale gave yellow crystals of **2**, crystallized chloroform – methanol (1:1), 225 mg, m. p. : 174 - 175 °C; UV λ_{max} (MeOH): 215 nm (log ϵ 2.1); IR υ_{max} (KBr): 3391, 2924, 2853, 1738, 1725, 1645, 1461, 1376, 1265, 1167, 1085, 833, 722 cm⁻¹; ¹H NMR (CDCl₃): δ 5.58 (1H, m, H-10'), 5.43 (1H, m, H-12'), 5.37 (2H, m, H-9', H-13'), 4.31 (1H, m, H-2), 4.16 (2H, d, J = 6.0 Hz, H₂-3), 4.04 (2H, d, J = 7.5 Hz, H₂-1), 2.32 (2H, m, H₂-11'), 2.21 (2H, t, J = 7.2 Hz, H₂-2'), 2.19 (2H, t, J = 7.5 Hz, H₂-2''), 2.17 (4H, m, H₂-8', H₂-8''), 2.04 (4H, m, H₂-14', H₂-11''), 1.62 (2H, m, CH₂), 1.60 (2H, m, CH₂), 1.47 (2H, m, CH₂), 1.30 (12H, brs, 6 x CH₂), 1.25 (20H, brs, 10 x CH₂), 0.89 (3H, t, J = 6.3 Hz, Me-18'), 0.84 (3H, t, J = 6.1 Hz, Me-18''); ¹³C NMR (CDCl₃): δ 171.63 (C-1'), 169.87 (C-1''), 147.95 (C-12'), 133.36 (C-10'), 130.09 (C-9'), 127.45 (C-13'), 125.29 (C-9''), 116.23 (C-10''), 71.65 (C-2), 68.98 (C-3), 62.18 (C-1), 59.52 (C-11'), 55.81 (C-2'), 53.63 (C-2''), 36.86 (CH₂), 35.37 (CH₂), 34.08 (CH₂), 33.97 (CH₂), 31.98 (CH₂), 31.90 (CH₂), 29.76 (6 x CH₂), 29.41 (4 x CH₂), 29.16 (2 x CH₂), 27.44 (CH₂), 27.25 (CH₂), 25.77 (CH₂), 24.87 (CH₂), 22.68 (CH₂), 14.16 (Me-18'), 14.55 (Me-18''); EIMS *m*/z (rel. int.): 698 [M]⁺ (C₃₉H₇₁O₈P) (2.3).

Glyceryl 1,2-dioleio-3-phosphate (3)

Further elution of the column with chloroform produced pale yellow crystals of **3**, crystallized from chloroform - methanol (1:1), 187 mg, m. p. : 181 - 182 °C; UV λ_{max} (MeOH): 213 nm (log ϵ 2.3); IR υ_{max} (KBr): 3410, 2928, 2852, 1721, 1712, 1648, 1461, 1378, 1267, 1169, 1072, 1025, 725 cm⁻¹; ¹H NMR (CDCl₃): δ 5.54 (1H, m, H-9'), 5.41 (1H, m, H-9''), 5.39 (1H, m, H-10'), 5.35 (1H, m, H-10''), 4.25 (1H, m, H-2), 4.17 (2H, d, J = 6.0 Hz, H₂-3), 4.08 (2H, d, J = 7.1 Hz, H₂-1), 2.34 (2H, t, J = 7.2 Hz, H₂-2'), 2.29 (2H, t, J = 6.8 Hz, H₂-2''), 2.21 (2H, m, H₂-8'), 2.19 (2H, m, H₂-8''), 2.05 (2H, m, H₂-11'), 2.02 (2H, m, H₂-11''), 1.67 (2H, m, CH₂), 1.61 (2H, m, CH₂), 1.53 (2H, m, CH₂), 1.47 (2H, m, CH₂), 1.30 (12H, brs, 6 x CH₂), 1.26 (24H, brs, 12 x CH₂), 0.88 (3H, t, J = 6.5 Hz, Me-18'), 0.83 (3H, t, J = 6.6 Hz, Me-18''); ¹³C NMR (CDCl₃): δ 169.83 (C-1'), 169.68 (C-1''), 131.44 (C-9'), 131.39 (C-9''), 125.29 (C-10'), 125.25 (C-10''), 83.16 (C-2), 74.58 (C-3), 62.19 (C-1), 36.83 (C-2'), 35.79 (C-2''), 31.91 (CH₂), 31.01 (CH₂), 29.77 (6 x CH₂), 29.42 (6 x CH₂), 29.17 (8 x CH₂), 27.83 (CH₂), 27.61 (CH₂), 22.69 (CH₂), 22.68 (CH₂), 14.18 (Me-18''); EIMS *m*/z (rel. int.): 700 [M]⁺ (C₃₉H₇₃O₈P) (8.9).

Gallic acid (4)

Elution of the column with chloroform - methanol (19:1, ν/ν) afforded a colorless amorphous powder of **4**, purified from chloroform - methanol (1:1, ν/ν), 314 mg, m. p. 235 - 238 °C; UV λ max (MeOH): 218, 269 nm (log ε 4.6, 1.5); IR γ_{max} (KBr): 3413, 2923, 2837, 1690, 1632, 1561, 1408, 1071, 908 cm⁻¹; ¹H NMR (CDCl₃): δ 12.33 (1H, brs, COOH), 7.28 (1H, d, J = 2.0 Hz, H-2), 6.83 (1H, d, J = 2.0 Hz, H-6); ¹³C NMR (CDCl₃): δ 137.42 (C-1), 127.52 (C-2), 140.68 (C-3), 150.38 (C-4), 140.47 (C-5), 125.81 (C-6), 183.12 (C-7); EI MS *m*/z (rel. int.): 170 [M]⁺ (C₇H₆O₅) (6.1).

(Z)- n-Hexatriacontan-12-en-1-ol (5)

Elution of the column with chloroform – methanol (49:1) furnished colourless crystals of **5**, 125 mg, m. p. 85 - 87 ° C; UV λmax (MeOH): 216 nm (log ε 3.6); IR γmax (KBr): 3431, 2921, 2849, 1633, 1451, 1388, 1266, 1125, 1031, 728 cm⁻¹; ¹H NMR (CDCl₃): δ 5.20 (1H, m, w_{1/2} = 7.9 Hz, H-11, H-12), 3.34 (2H, t, J = 7.8 Hz, H₂-1), 2.16 (2H, m, H₂-10), 2.01 (2H, m, H₂-13), 1.25 (60H, brs, 30 × CH₂), 0.86 (3H, t, J = 6.4 Hz, Me-36); ¹³C NMR (CDCl₃): δ 137.79 (C-11), 130.05 (C-12), 60.16 (C-1), 49.15 (C-10), 38.93 (C-13), 30.46 (CH₂), 29.34 (8 x CH₂), 29.28 (11 x CH₂), 29.05 (5 x CH₂), 28.33 (CH₂), 27.23 (CH₂), 25.81 (CH₂), 24.78 (CH₂), 22.69 (CH₂), 14.39 (Me-36); EIMS *m*/z (rel. int.): 520 [M]⁺ (C₃₆H₇₂O) (19.1), 363 (7.2), 337 (5.8), 183 (11.2), 157 (8.2).



6β-Hydroxylupdienolyl 3β-oleate (6)

Further elution of the column with chloroform-methanol (49:1) offered colourless crystals of **6**, recrystallized from acetone, 143 mg, m. p. 134 - 135 °C; UV λ_{max} (MeOH): 223 nm (log ϵ 3.5); IR γ_{max} (KBr): 3449, 2912, 2857, 1731, 1645, 1458, 1379, 1247, 1027, 754 cm⁻¹; ¹H NMR (DMSO-d₆): δ 5.34 (1H, t, J = 5.2 Hz, H-12), 5.13 (1H, m, H-9'), 5.11 (1H, m, H-10'), 4.68 (1H, s, H₂-29a), 4.66 (1H, s, H₂-29b), 4.50 (1H, dd, J = 4.8, 7.6 Hz, H-3\alpha), 4.16 (1H, ddd, J = 2.0, 1.6, 8.2 Hz, H-6\alpha), 2.41 (2H, d, J = 8.1 Hz, H-18\beta), 2.33 (2H, t, J = 8.1 Hz, H₂-2'), 2.23 (1H, m, H-19), 2.08 (1H, m, H₂-11), 2.01 (1H, m, H₂-8'), 1.91 (1H, m, H₂-11'), 1.68 (3H, brs, Me-30), 1.07 (3H, brs, Me-23), 1.04 (3H, brs, Me-26), 0.90 (3H, brs, Me-25), 0.87 (3H, brs, Me-24), 0.85 (3H, s, Me-27), 0.82 (3H, t, J = 6.3 Hz, Me-18'), 0.79 (3H, s, Me-28), 1.65 - 1.13 (48H, m, 4 x CH, 22 x CH₂); ¹³C NMR (DMSO-d₆): δ 40.89 (C-1), 23.42 (C-2), 81.02 (C-3), 38.51 (C-4), 55.31 (C-5), 72.68 (C-6), 34.26 (C-7), 41.59 (C-8), 50.39 (C-9), 37.76 (C-10), 21.46 (C-11), 124.37 (C-12), 139.88 (C-13), 42.36 (C-14), 29.21 (C-15), 28.13 (C-16), 47.69 (C-17), 59.10 (C-18), 48.05 (C-19), 150.98 (C-20), 21.30 (C-21), 33.82 (C-22), 28.01 (C-23), 22.75 (C-24), 21.46 (C-25), 16.23 (C-26), 15.79 (C-27), 18.06 (C-28), 109.43 (C-29), 19.35 (C-30), 171.10 (C-1'), 43.05 (C-2'), 36.84 (C-3'), 34.47 (C-4'), 29.76 (C-5'), 29.53 (C-6'), 29.43 (C-7'), 42.87 (C-8'), 121.75 (C-9'), 118.95 (C-10'), 42.12 (C-11'), 29.32 (C-12'), 26.65 (C-13'), 25.14 (C-14'), 23.75 (C-15'), 22.60 (C-16'), 21.99 (C-17'), 14.19 (C-18'); EIMS *m*/z (rel.int.): 704 [M]⁺ (C₄₈H₈₀O₃) (41.3), 488 (33.2), 439 (29.6), 281 (6.3), 265 (23.5), 224 (11.7), 216 (15.8).

Stigmast-5-en-6α-ol-26-oic acid 3β-linolenate (7)

Elution of the column with chloroform - methanol (19:1, ν/ν) furnished a pale yellow amorphous powder of **7**, recrystallized from chloroform – methanol (1:1), 163 mg; m. p. 90 -91 °C; UV λ max (MeOH): 232 nm (log ϵ 2.9); IR γ_{max} (KBr): 3429, 3201, 2925, 2853, 1729, 1703, 1645, 1601, 1462, 1379, 1272, 1123, 1072, 1040, 981, 741 cm⁻¹; ¹H NMR (CDCl₃): δ 5.35 (1H, d, J = 5.1 Hz, H- 6), 5.32 (2H, m, H- 10', H-12'), 5.29 (1H, m, H- 13'), 5.27 (3H, m, H- 9', H-15', H-16'), 4.21 (1H, brs, w $_{\nu_2}$ = 18.1 Hz, H- 3 α), 4.07 (1H, d, J = 6.8 Hz, H-4 β), 2.78 (2H, m, H₂-11'), 2.53 (2H, m, H₂-14), 2.34 (2H, t, J = 7.2 Hz, H₂-2'), 2.29 (1H, m, H-25), 2.03 – 1,07 (37H, m, 16 x CH₂, 5 x CH), 1.02 (3H, brs, Me-19), 0.92 (3H, d, J = 6.3 Hz, Me- 21), 0.87 (3H, d, J = 6.1 Hz, Me- 27), 0.84 (3H, t, J = 6.6 Hz, Me-18'), 0.80 (3H, t, J = 7.2 Hz, Me- 29), 0.68 (3H, brs, Me-18); ¹³C NMR (CDCl₃): δ 37.32 (C-1), 31.89 (C-2), 71.88 (C-3), 68.23 (C-4), 140.79 (C-5), 121.78 (C-6), 29.31 (C-7), 34.43 (C-8), 50.23 (C-9), 36.24 (C-10), 23.16 (C-11), 38.83 (C-12), 39.85 (C-13), 56.83 (C-14), 27.23 (C-15), 28.21 (C-16), 56.13 (C-17), 11.76 (C-18), 19.46 (C-19), 36.71 (C-20), 19.11 (C-21), 33.96 (C-22), 26.21 (C-23), 45.89 (C-24), 29.63 (C-25), 179.20 (C-26), 19.76 (C-27), 24.93 (C-28), 11.96 (C-29), 169.25 (C-1'), 39.77 (C-2'), 29.96 (C-3'), 29.76 (C-4'), 29.71 (C-5'), 29.60 (C-6'), 29.67 (C-7'), 29.26 (C-8'), 132.50 (C-9'), 131.68 (C-10'), 35.58 (C-11'), 129.43 (C-12'), 129.16 (C-13'), 38.18 (C-14'), 128.61 (C-15'), 127.16 (C-16'), 22.67 (C-17'), 17.16 (C-18'); EIMS *m*/z (rel. int.): 720 [M]⁺ (C₄₇H₇₆O₅) (49.8), 459 (13.1), 442 (25.6), 288 (29.8), 278 (23.2), 271 (26.9), 261 (22.6), 256 (23.1), 253 (21.8), 214 (20.1), 171 (3.5).

Glucuronosyl-(6→1′)-glucoside (8)

Elution of the column with chloroform - methanol (9 : 1) yielded colourless crystals of **8**, recrystallized from chloroform-methanol (1 : 1), 127 mg; UV λ_{max} (MeOH): 211 nm (log ϵ 4.2); IR γ_{max} (KBr): 3420, 3395, 3260, 2924, 2854, 1726, 1627, 1352, 1047 cm⁻¹; ¹H NMR (DMSO-d₆): δ 5.03 (1H, d, J = 7.2 Hz, H-1), 4.84 (1H, m, H-5), 3.96 (1H, m, H-2), 3.74 (1H, m, H-3), 3.63 (1H, m, H-4); 4.87 (1H, d, J = 7.3 Hz, H-1'), 4.13 (1H, m, H-5'), 3.81 (1H, m, H-2'), 3.72 (1H, m, H-3'), 3.69 (1H, m, H-4'), 3.21 (2H, d, J = 9.0 Hz, H₂-6'); ¹³C NMR (DMSO-d₆); δ 104.39 (C-1), 70.82 (C-2), 68.54 (C-3), 74.63 (C-4), 77.19 (C-5), 171.36 (C-6), 101.41 (C-1'), 72.89 (C-2'), 70.51 (C-3'), 67.10 (C-4'), 75.74 (C-5'), 61.71 (C-6'); EI MS *m*/z (rel. int.): 356 [M]⁺ (C₁₂H₂₀O₁₂) (34.3), 193 (12.9), 179 (8.5), 163 (14.1).

Isolation of phytoconstituents from the stem bark of Ficus religiosa

Arachidic acid (9)

Elution of the column with petroleum ether gave colorless amorphous powder of **9**, purified from chloroformmethanol (1:1, ν/ν), 203 mg, m. p. 74 - 75 °C; R_f 0.33 (acetone-methanol, 9:1); UV λ_{max} (MeOH): 206, 223 nm (log ϵ



3.7, 1.8). IR γ_{max} (KBr): 3422, 2925, 2854, 1698, 1456, 1368, 1218, 1166, 1030, 728 cm⁻¹; ¹H NMR (CDCl₃): δ 2.74 (2H, t, J = 7.2 Hz, H₂-2), 2.19 (2H, m, CH₂), 1.58 (2H, brs, CH₂), 1.25 (30 H, brs, 15 x CH₂), 0.84 (3H, t, J = 6.8 Hz, Me-20). ¹³C NMR (CDCl₃): δ 178.13 (C-1), 52.48 (C-2), 30.43 (CH₂), 28.65 (14 x CH₂), 28.65 (CH₂), 22.34 (CH₂), 13.62 (Me-20). EI MS *m*/z (rel.int.): 312 [M]⁺ (C₂₀H₄₀O₂) (43.8).

Glyceryl-1-oleiyl-2-palmityl-3-stearate (10)

Further elution of the column with petroleum ether furnished a pale yellow viscous mass of **10**, purified by preparative TLC using petroleum ether - chloroform (3:1), UV λ_{max} (MeOH): 213 nm (log ϵ 2.3); IR γ_{max} (KBr): 2922, 2853, 1734, 1721, 1638, 1463, 1376, 1272, 1246, 1173, 1121, 1073, 1030, 905, 725 cm⁻¹; ¹H NMR (CDCl₃): δ 5.36 (1H, m, H-9'), 5.33 (1H, m, H-10'), 4.21 (1H, m, H-2), 4.13 (2H, m, H₂-1), 4.09 (2H, d, J = 6.8 Hz, H₂-3), 2.38 (2H, t, J = 7.6 Hz, H₂-2'), 2.31 (2H, t, J = 7.4 Hz, H₂-2''), 2.28 (2H, t, J = 7.3 Hz, H₂-2'''), 2.07 (2H, m, H₂-8'), 1.65 (2H, m, H₂-11'), 1.55 (2H, m, 2 x CH₂), 1.30 (6H, m, 3 x CH₂), 1.28 (20H, brs, 10 x CH₂), 1.23 (48H, brs, 24 x CH₂), 0.88 (3H, t, J = 7.2 Hz, Me-18'), 0.84 (3H, t, J = 6.9 Hz, Me-16''), 0.79 (3H, t, J = 6.1 Hz, Me-16'''); ¹³C NMR (CDCl₃): δ 172.6 (C-1'), 169.43 (C-1''), 168.75 (C-1'''), 131.96 (C-9'), 128.08 (C-10'), 69.21 (C-2), 65.16 (C-1), 62.34 (C-3), 41.35 (C-2') 36.08 - 29.85 (10 x CH₂), 29.64 (7 x CH₂), 29.61 - 29.03 (14 x CH₂), 28.89 (CH₂), 27.66 (CH₂), 26.91 (2 x CH₂), 25.83 (4 x CH₂), 24.72 (CH₂), 22.68 (3 x CH₂), 14.23 (Me-18'), 14.12 (Me-16''), 13.41 (Me-18'''); EI MS *m*/z (rel. int.): 860 [M]⁺ (C₅₅H₁₀₄O₆) (2.3), 265 (8.2), 239 (11.8).

1,2,3-Trioleiyl β-D-arabinoside (11)

Elution of the column with chloroform afforded pale yellow amorphous powder of 11, recrystallized from chloroform-methanol (1:1), 219 mg; m. p. 115 -117 °C; UV λ_{max} (MeOH): 244 nm (log ε 4.3); IR γ_{max} (KBr): 3447, 2918, 2823, 1739, 1725, 1645, 1456, 1377, 1244, 1172, 1095, 1029, 837, 758 cm⁻¹; ¹H NMR (DMSO-d₆): δ 5.42 (1H, d, J = 7.1 Hz, H-1), 4.30 (1H, m, H-2), 4.11 (1H, m, H-3), 3.81 (1H, m, H-4), 3.52 (2H, m, H₂-5), 5.35 (2H, m, H-9', H-9''), 5.21 (1H, m, H-9'''), 5.17 (1H, m, H-10'), 5.11 (1H, m, H-10''), 5.06 (1H, m, H-10'''), 2.30 (2H, t, J = 7.1 Hz, H₂-2'), 2.27 (4H, m, H₂-2", H-2"'), 2.15 (6H, m, H₂-8", H₂-8", H₂-8"'), 2.02 (6H, m, H₂-11', H₂-11", H₂11"'), 1.66 (6H, m, H₂-3', H-3", H-3"), 1.59 (6H, m, 3 x CH₂), 1.29 (20H, brs, 10 x CH₂), 1.25 (36H, brs, 18 x CH₂), 1.22 (16H, brs, 8 x CH₂), 0.87 (3H, t, J = 6.3 Hz, CH₃-18'), 0.84 (3H, t, J = 6.1 Hz, CH₃-18"), 0.80 (3H, t, J = 5.9 Hz, CH₃-18"'); ¹³C NMR (DMSO-d₆): δ 109.13 (C-1), 80.01 (C-2), 70.12 (C-3), 68.31 (C-4), 61.42 (C-5), 174.73 (C-1'), 172.22 (C-1"), 169.79 (C-1""), 134.23 (C-9'), 130.59 (C-9"), 129.45 (C-9""), 125.07 (C-10"), 124.47 (C-10"), 123.74 (C-10"), 59.33 (C-2'), 58.38 (C-2"), 54.66 (C-2""), 47.01 (C-8"), 41.43 (C-8"), 40.91 (C-8""), 36.18 (C-11'), 34.95 (C-11"), 33.60 (C-11""), 34.95 (CH₂), 33.46 (CH₂), 33.29 (CH₂), 33.15 (CH₂), 32.21 (CH₂), 31.58 (CH₂), 31.29 (CH₂), 33.12 (CH₂), 30.85 (CH₂), 29.05 (6 x CH₂), 28.83 (CH₂), 28.70 (CH₂), 28.55 (CH₂), 27.58 (CH₂), 27.46 (CH₂), 26.77 (CH₂), 26.55 (CH₂), 25.98 (CH₂), 25.79 (CH₂), 25.15 (CH₂), 25.02 (CH₂), 24.91 (CH₂), 24.36 (CH₂), 22.47 (2 x CH₂), 22.03 (CH₂), 15.18 (Me-18'), 13.79 (Me-18''), 13.66 (Me-18''); EIMS m/z (rel. int.): 942 [M]⁺ $(C_{59}H_{106}O_8)$ (2.3), 281 (11.3), 265 (16.1), 147 (8.6).

Glyceryl 1-oleio-2-stearyl-3-phosphate (12)

Further elution of the column with chloroform produced pale yellow crystals of **12**, crystallized from chloroform - methanol (1:1), 187 mg, m. p. : 99 - 101 °C; UV λ_{max} (MeOH): 215 nm (log ϵ 2.8); IR γ_{max} (KBr): 3435, 2923, 2853, 1736, 1722, 1640, 1458, 1376, 1244, 1173, 1030, 837, 723 cm⁻¹; ¹H NMR (CDCl₃): δ 5.31 (1H, m, H-9'), 5.22 (1H, m, H-10'), 4.13 (1H, m, H-2), 4.08 (2H, m, H₂-3), 4.03 (2H, m, H₂-1), 2.27 (2H, t, J = 7.1 Hz, H₂-2'), 2.13 (2H, m, H₂-2''), 2.03 (2H, m, H₂-8'), 1.98 (2H, m, H₂-11'), 1.65 (2H, m, CH₂), 1.58 (2H, m, CH₂), 1.25 (56H, brs, 26 x CH₂), 0.95 (3H, t, J = 6.6 Hz, Me-18'), 0.87 (3H, t, J = 6.3 Hz, Me-18''); ¹³C NMR (CDCl₃): δ 171.16 (C-1'), 170.29 (C-1''), 134.45 (C-9'), 124.43 (C-10'), 81.26 (C-2), 71.06 (C-3), 61.58 (C-1), 59.47 (C-2'), 55.39 (C-2''), 50.35 (CH₂), 46.61 (CH₂), 34.84 (CH₂), 33.61 (CH₂), 31.58 (CH₂), 31.28 (CH₂), 30.63 (CH₂), 29.05 (10 x CH₂), 28.72 (CH₂), 28.51 (CH₂), 27.46 (CH₂), 26.57 (CH₂), 25.78 (CH₂), 25.14 (CH₂), 25.01 (CH₂), 24.35 (CH₂), 22.87 (CH₂), 22.05 (CH₂), 17.36 (Me-18'), 13.57 (Me-18''); EIMS *m*/z (rel. int.): 702 [M]⁺ (C₃₉H₇₅O₈P) (4.8).



Glyceryl 1, 2-distearyl-3-phosphate (13)

Further elution of the column with chloroform pale yellow crystals of **13**, recrystallized from chloroform - methanol (1:1), 218 mg, m. p. : 121 - 122 °C; UV λ_{max} (MeOH): 217 nm (log ϵ 2.1); IR γ_{max} (KBr): 3402, 2930, 2856, 1725, 1721, 1642, 1455, 1373, 1226, 1187, 1033, 757 cm⁻¹; ¹H NMR (CDCl₃): δ 4.23 (1H, m, H-2), 4.11 (2H, m, H₂-3), 4.04 (2H, m, H₂-1), 2.65 (2H, m, H₂-2'), 2.26 (2H, m, H₂-2''), 1.98 (2H, m, CH₂), 1.65 (2H, m, CH₂), 1.56 (2H, m, CH₂), 1.27 (56H, brs, 28 x CH₂), 0.87 (3H, t, J = 6.3 Hz, Me-18'), 0.84 (3H, t, J = 6.1 Hz, Me-18''); ¹³C NMR (CDCl₃): δ 173.12 (C-1'), 171.35 (C-1''), 72.13 (C-2), 65.86 (C-3), 63.05 (C-1), 31.97 (CH₂), 30.99 (CH₂), 29.75 (27 x CH₂), 29.41 (CH₂), 27.24 (CH₂), 25.29 (CH₂), 22.74 (CH₂), 14.17 (Me-18'), 14.12 (Me-18''); EI MS *m*/z (rel. int.): 704 [M]⁺ (C₃₉H₇₇O₈P) (8.9).

Glyceryl 1-linoleiyl-2-arachidyl-3-phosphate (14)

Further elution of the column with chloroform offered pale yellow crystals of **14**, crystallized from chloroform - methanol (1:1), 132 mg, m. p. : 168 - 170 °C; UV λ_{max} (MeOH): 216 nm (log ε 3.2); IR γ_{max} (KBr): 3411, 2925, 2852, 1721, 1643, 1461, 1377, 1246, 1167, 1043, 758 cm⁻¹; ¹H NMR (CDCl₃): δ 5.36 (1H, m, H-9'), 5.34 (1H, m, H-10'), 5.12 (2H, m, H-12', H-13'), 4.14 (1H, m, H-2), 4.07 (2H, d, J = 7.2 Hz, H₂-3), 4.02 (2H, m, H₂-1), 2.31 (2H, t, J = 7.1 Hz, H₂-2'), 2.26 (2H, d, J = 7.2 Hz, H₂-2''), 2.04 (2H, m, H₂-11'), 1.97 (2H, m, H₂-8'), 1.68 (2H, m, H₂-14'), 1.54 (2H, m, CH₂), 1.25 (34H, brs, 17 x CH₂), 1.22 (4H, m, 2 x CH₂), 0.89 (3H, t, J = 6.3 Hz, Me-18'), 0.84 (3H, t, J = 6.1 Hz, Me-20''); ¹³C NMR (CDCl₃): δ 171.13 (C-1'), 169.28 (C-1''), 131.65 (C-9'), 127.86 (C-10'), 125.73 (C-12'), 122.58 (C-13'), 82.38 (C-2), 75.93 (C-3), 62.54 (C-1), 55.88 (C-11'), 55.06 (C-2'), 47.18 (C-2''), 39.67 (CH₂), 34.11 (CH₂), 31.98 (CH₂), 29.75 (19 x CH₂), 29.41 (CH₂), 29.16 (CH₂), 27.25 (CH₂), 24.74 (CH₂), 22.74 (CH₂), 14.20 (Me-18'), 14.18 (Me-20''); EI MS *m*/z (rel. int.): 728 [M]⁺ (C₄₁H₇₇O₈P) (3.7).

2-(2'-oleiyl glucosyl) gallic acid (15)

Elution of the column with chloroform - methanol (19:1) afforded a pale yellow amorphous powder of **15**, recrystallized from chloroform-methanol (1:1), 1.2 g, m. p.: 117 – 118 °C; UV λ_{max} (MeOH): 297 nm (log ϵ 4.1); IR γ_{max} (KBr): 3510, 3435, 3365, 2924, 2853, 1725, 1703, 1636, 1541, 1460, 1385, 1218, 1046, 769 cm⁻¹; ¹H NMR (CDCl₃): δ 7.43 (1H, d, J = 2.9 Hz, H-2), 7.27 (1H, d, J = 2.9 Hz, H-6), 5.12 (1H, d, J = 7.1 Hz, H-1'), 4.37 (1H, m, H-5'), 4.13 (1H, m, H-2'), 3.86 (1H, m, H-3'), 3.78 (1H, m, H-4'), 3.19 (2H, d, J = 9.5 Hz, H₂-6'), 5.35 (2H, m, H-9", H-10"), 2.38 (2H, t, J = 7.2 Hz, H₂-2"), 2.11 (2H, m, H₂-8'), 2.03 (2H, m, H₂-11'), 1.54 (2H, m, CH₂), 1.24 (20H, brs, 10 x CH₂), 0.94 (3H, t, J = 6.3 Hz, Me-18"); ¹³C NMR (CDCl₃): δ 139.25 (C-1), 127.81 (C-2), 160.06 (C-3), 152.76 (C-4), 140.82 (C-5), 114.07 (C-6), 180.32 (C-7), 103.23 (C-1'), 74.36 (C-2'), 71.63 (C-3'), 68.54 (C-4'), 77.63 (C-5'), 61.73 (C-6'), 171.64 (C-1"), 57.40 (C-2"), 29.73 (C-3"), 29.59 (C-4"), 29.75 (C-13"), 28.41 (C-14"'), 27.09 (C-15"), 25.81 (C-16"), 22.69 (C-17"), 14.17 (C-18"); EI MS *m*/z (rel. int.): 596 [M]⁺ (C₃₁H₄₈O₁₁) (4.3).

Results and Discussion

Compounds 1 - 3 were the known lipid constituents characterized as glyceryl 1-oleio-2,3-distearate (glyceryl linolyldistearin, 1), glyceryl 1- linolyl-2-oleio-3-phosphate (2) and glyceryl 1,2-dioleio-3-phosphate (3) [33 - 35]. Compound 4 was a known aromatic acid identified as gallic acid [36,37].

Compound **5** had IR absorption bands for hydroxyl group (3431 cm⁻¹), unsaturation (1633 cm⁻¹), and long aliphatic chain (728 cm⁻¹). Its mass spectrum exhibited a molecular ion peak at m/z 520 corresponding to an aliphatic alcohol, C₃₆H₇₂O. The prominent ion peaks arising at m/z 337 [C₁₂ – C₁₃ fission, CH₃-(CH₂)₂₃]⁺, 183 [M – 337]⁺, 363 [C₁₀ – C₁₁ fission, CH₃-(CH₂)₂₃-CH=CH]⁺ and 157 [M - 363]⁺ indicated the existence of the hydroxyl group at terminal C₁ carbon and vinylic carbon at C-11 position. The ¹H NMR spectrum of **5** showed a two - proton multiplet at δ 5.20 with half-width of 7.9 Hz assigned to cis-oriented vinylic H-11 and H-12 protons. A two – proton triplet at δ 3.34 (J = 7.8 Hz) was accounted to the primary hydroxymethylene H₂-1 protons. The other methylene protons appeared as two-proton multiplets at δ 2.16 and 2.01 and as broad singlet at δ 1.25 (60 H). A three-proton triplet at δ 0.86 (J = 6.4 Hz) was attributed to terminal C-36 primary methyl protons. The ¹³C NMR spectrum of **5** displayed signals for



vinylic carbons at δ 137.79 (C-11) and 130.05 (C-12), hydroxymethylene carbon δ 60.16 (C-1), other methylene carbons from δ 49.15 to 22.69 and methyl carbon at δ 14.39 (C-36). On the basis of above discussion the structure of **5** was established as (Z)-*n*-hexatriacont-12-en-1-ol, a new aliphatic alcohol.

Compound **6**, named 6 β -hydroxylupdienolyl 3 β -oleate, showed distinctive IR absorption bands for hydroxyl group (3449 cm⁻¹), ester function (1731 cm⁻¹), unsaturation (1645 cm⁻¹) and long aliphatic chain (754 cm⁻¹). Its molecular ion peak was determined on the basis of mass and ¹³C NMR spectra at m/z 704 consistent with a molecular formula of a triterpenic ester, C₄₈H₈₀O₃. The prominent ion peaks generated at m/z 488 [C₃₂H₅₆O₃]⁺ and 216 [C₁₆H₂₄]⁺ due to retro-Diels-Alder fragmentation pattern suggested location of one of the vinylic linkage at C-12 in ring C, another vinylic linkage of in the ring D/E and location of the ester linkage and hydroxyl group in rings A and B, respectively. The ion fragments produced at m/z 265 [CH₃-(CH₂)₇-CH=CH-(CH₂)₇CO]⁺, 281 [CH₃-(CH₂)₇-CH=CH-(CH₂)₇COO]⁺ and 224 [478 - 265]⁺ indicated that oleic acid was esterified with the triterpenol.

The ¹H NMR spectrum of **6** showed a one-proton triplet at δ 5.34 (J = 5.2 Hz) and two one - proton multiplets at δ 5.13 and 5.11 assigned to vinylic H-12, H - 9' and H - 10' protons, respectively, two one-proton singlets at δ 4.68 and 4.66 ascribed to unsaturated methylene H₂ -29 protons of a lupene-type triterpene and two one - proton signals as a double doublet at δ 4.50 (J = 4.8, 7.6 Hz) and as a triple doublet 4.16 (J = 2.0, 1.6, 8.2 Hz) correspondingly accounted to α -oriented oxymethine H-3 and carbinol H-6 α protons. A three - proton singlet in the deshielded region at δ 1.68 was attributed to C-30 methyl protons located on C-20 vinylic carbon. Six three - proton singlets between 1.07 - 0.79 were associated with the tertiary C-23 to C-28 methyl protons. A three - proton triplet at δ 0.82 (J = 6.3 Hz) with coupling interaction of 6.3 Hz was accommodated in the primary C-18' methyl protons. A two - proton triplet at δ 2.33 (J = 8.1 Hz) was due to methylene H₂-2' protons adjacent to the ester group. The signals between δ 2.41 - 1.13 were due to the remaining methylene and methine protons. The ¹³C NMR spectrum of **6** displayed signals for ester carbon at δ 171.10 (C-1'), vinylic carbons at δ 150.98 (C-20), 109.43 (C-29), 121.75 (C-9') and 118.95 (C-10'), oxymethine carbons at δ 81.02 (C-3) and 72.68 (C-6) and methyl carbons between δ 28.01 - 14.19. The ¹H and ¹³C NMR spectral values of the triterpenic unit were compared with related lupene-type molecules [38-40]. On the basis of spectral data analysis and chemical reactions, the structure of **6** was formulated as lup-12,20(29)-dien-6\beta-ol-3\beta-olyl oleate, a new lupene-type triterpenic ester.

Compound **7**, named stigmast-5-en- 6α -ol-26-oic acid 3β -linolenate, gave effervescences with sodium bicarbonate solution and exhibited characteristic IR absorption bands for hydroxyl function (3429 cm^{-1}), ester group (1703 cm^{-1}), unsaturation (1645, 1601 cm^{-1}) and long aliphatic chain (741 cm^{-1}). Its molecular ion peak was determined at m/z 720 on the basis of mass and ¹³C NMR spectra consistent with a molecular formula of a sterol ester $C_{47}H_{76}O_5$. The ion peaks arising at m/z 261 [$C_{1'}$ - O fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [C_3 - O fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [C_3 - O fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_3CO$]⁺, 459 [M - 261]⁺, 278 [$C_3 - O$ fission, $CH_3(CH_2)_7$ ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$) ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$) ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$) ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$ ($CH=CHCH_2)_7$ (CH=CH

The ¹H NMR spectrum of **7** displayed a one-proton doublet at δ 5.35 (J=5.1 Hz) assigned to vinylic H-6 proton. Three multiplets at δ 5.32 (2H), 5.29 (1H) and 5.27 (3H) were ascribed to vinylic protons of the linolenate ester chain. A one-proton broad multiplet at δ 4.21 with half-width of 18.1 Hz and a one-proton doublet at δ 4.07 (J = 6.8 Hz) were ascribed to α -oriented oxygenated methine H-3 and β -oriented carbinol H-4 protons, respectively. A two-proton triplet at δ 2.34 (J=7.6 Hz) was due to methylene H₂-2' protons adjacent to the ester function. Two three-proton broad singlets at δ 1.02 and 0.68, two three- protons doublets at δ 0.92 (J=6.3 Hz) and 0.87 (J=6.1 Hz) and two three-proton triplets at δ 0.84 (J=6.6 Hz) and 0.80 (J= 7.2 Hz) were associated with tertiary C-19 and C-18, secondary C-21 and C-27 and primary C-18' and C-29 methyl protons, respectively, all attached to the saturated carbons. The remaining methylene and methine protons resonated from δ 2.78 to 1.07. The ¹³C NMR spectrum of **7** displayed signals for carboxylic carbons at δ 179.20 (C-26), ester carbon at δ 169.25 (C-1'), vinylic carbons between δ 140.7 - 121.78, oxymethine carbon at δ 71.88 (C-3), carbinol carbon at δ 68.23 (C-4) and methyl carbons from at δ



19.76 to 11.76. The ¹H and ¹³C NMR spectral data of steroidal nucleus were compared with the reported spectral values of steroids [41-43]. On the basis these evidences the structure of **7** has been elucidated as stigmast-5-en-4 α -ol-26-oic acid-3 β -olyl linolenate, a new steroidal ester.

Compound **8**, designated as glucuronosyl-($6 \rightarrow 1'$)-glucoside, gave positive tests for glycosides and displayed characteristic IR absorption bands for hydroxyl groups (3420, 3395, 3260 cm⁻¹) and ester function (1726 cm⁻¹). Its mass spectrum exhibited a molecular ion peak at m/z 356 corresponding to a molecular formula of a diglycoside, C ${}_{12}H_{20}O_{12}$. The ion fragments generating at m/z 193 [C₆H₉O₇]⁺, 179 [C₆H₁₁O₆]⁺ and 163 [C₆H₁₁O₅]⁺ indicated that a hexose unit was linked with C₆ sugar acid. The ¹H NMR spectrum of **8** exhibited two one-proton doublets at δ 5.03 (J = 7.2 Hz) and δ 4.87 (J = 7.3 Hz) assigned to anomeric H-1 and H-1' protons, respectively. Eight one-protons multiplets between δ 4.84 - 3.69 were ascribed to carbinol protons of the sugar unit. A two-proton doublet at δ 3.21 (J = 9.0 Hz) was accounted to hydroxymethylene H₂-6' protons. The ¹³C NMR spectrum of **8** exhibited signals for the ester carbon at δ 171.36 (C-6), anomeric carbons at δ 104.39 (C-1) and δ 101.41 (C-1') and other sugar carbons in the range from δ 77.19 to 61.36. The formation of the ester linkage between glucose and glucuronic acid was supported by the IR absorption band at 1726 cm⁻¹ and ¹³C NMR carbon signal at δ 171.36 indicating the attachment of one sugar unit with another sugar by a ($6 \rightarrow 1'$) linkage. Acid hydrolysis of **3** yielded D-glucose, R_f 0.12 and D-glucuronic acid, R_f 0.16 (*n*-butanol-acetic acid-water, 4:1:5). On the basis of these evidences, the structure of **8** has been elucidated as β -D-glucuropyranosyl-($6 \rightarrow 1'$)-O- β -D-glucopyranoside.



Glyceryl linolyl distearate (1)



Glyceryl 1-linolyl-2-oleio-3-phosphate (2)



Glyceryl 1,2-dioleio-3-phosphate (3)



Gallic acid (**4**) ³⁶ 12 11 1 CH₃(CH₂)₂₃CH=CH(CH₂)₈CH₂-CH₂OH (Z)-n-Hexatriacontan-11-en-1-ol (**5**)





Stigmast-5-en- 6α -ol-26-oic acid 3β -linolenate (7)



Glucuronosyl 6-O-glucoside (8)

Figure 1: Structural formulae of the compounds 1 - 8 isolated from the stem bark of Ficus racemosa

Compound **9** was a known fatty acid identified as *n*-eicosanoic acid (arachidic acid) [44]. Compound **10** was a mixed glyceride characterized as glyceryl-1-oleiyl-2-palmityl-3-stearate (glyceryl-1-octadec-9'-enoyl- 2-hexadecanoate-3-octadecanoate).

Compound **11**, named 1,2,3-trioleiyl β -D-arabinoside, decolourized bromine water and responded positively to glycoside tests. Its IR spectrum showed characteristic absorption bands for a hydroxyl group (3447 cm⁻¹), ester groups (1739, 1725 cm⁻¹), unsaturation (1645 cm⁻¹) and long aliphatic chain (758 cm⁻¹). On the basis of mass and ¹³C NMR spectra the molecular ion peak of **11** was determined at *m*/z 942 corresponding to a molecular weight of triacyl pentose, C₅₉H₁₀₆O₈. The generation of the prominent ion peaks at *m*/z 265 [C₁' - O fission, OC-(CH₂)₇-CH=CH-(CH₂)₇CH₃]⁺, 281 [C₁ - O fission, OOC-(CH₂)₇-CH=CH-(CH₂)₇CH₃]⁺ and 147 [M – 3 x 265, C₅H₇O₅]⁺ indicated that three units of oleic acid were esterified with a pentose sugar moiety.

The ¹H NMR spectrum of **11** displayed a one-proton doublet at δ 5.42 (J = 7.1 Hz) assigned to anomeric H-1 proton. The other sugar protons appeared as multiplets from δ 4.30 to 3.52. The presence of the signals as multiplets in the deshielded region between δ 5.35 – 5.06 were attributed to the vinylic protons of the acyl chains. The methylene protons resonated as multiplets in the range of δ 2.30 – 1.59 and as broad singlets at δ 1.29 (20H), 1.25 (36H) and 1.22 (16H). Three three-proton triplets at δ 0.87 (J = 6.3 Hz), 0.84 (J = 6.1 Hz) and 0.80 (J = 5.9 Hz) were accounted to terminal C-18', C-18'' and C-18''' primary methyl protons, respectively. The presence of the sugar protons in the deshielded region as a doublet at δ 5.42 (J = 7.1 Hz, H-1) and as multiplets at δ 4.30 (H-2) and 4.11 (H-3) indicated the attachment of oleiyl units at C-1, C-2 and C-3 positions, respectively. The ¹³C NMR spectrum of **11** showed



important signals for anomeric carbon at δ 109.13 (C-1), other sugar carbons between δ 80.01 - 61.42, ester carbons at δ 174.73 (C-1'), 172.22 (C-1") and 169.79 (C-1"'), vinylic carbons between δ 134.23 - 123.74, methylene carbons in range of δ 59.33 - 22.03 and methyl carbons at δ 15.18 (C-18'), 13.79 (C-18") and 13.66 (C-18"'). Acid hydrolysis of **11** yielded oleic acid, R_f 0.46 (acetic acid, 85%) and the sugar D-arabinose, R_f 0.70 (*n*-butonal : acetic acid: water, 4:1:1.6). On the basis of spectral data analysis and chemical reactions, the structure of **11** has been characterized as β -D-arabinopyranosyl 1,2,3-tri-octadec-9-enoate, a new triacyl arabinose.

Compounds **12**, **13** and **14** were the known phytoconstituents characterized as glyceryl 1-oleio-2-stearyl-3-phosphate, glyceryl 1,2-distearyl-3-phosphate and glyceryl 1-linoleiyl-2-arachidyl-3-phosphate [33-35], respectively.

Compound **15**, designated as 2-(2'-oleiyl glucosyl) gallic acid, responded for glycosidic and phenolic tests positively, had UV absorption maximum at 297 nm for aromaticity and IR absorption bands for hydroxyl groups (3510, 3435, 3365 cm⁻¹), ester function (1725 cm⁻¹), carboxylic group (1703 cm⁻¹), aromaticity (1636, 1541, 1046 cm⁻¹) and long aliphatic chain (769 cm⁻¹). On the basis of mass and ¹³C NMR spectral data, the molecular ion peak of **15** was determined at m/z 596 consistent with a molecular formula of a phenolic acid glycoside, C₃₁H₄₈O₁₁.

²⁰ CH₃(CH₂)₁₇CH₂-COOH



Glyceryl-1-oleiyl-2-palmityl-3-stearate (10)



Glyceryl-1-oleiyl-2-palmityl-3-phosphate (12)



Glyceryl-1-linoleiyl-2-arachidyl-3-phosphate (14)



1,2,3-Trioleiyl β -D-arabinose (11)



Glyceryl-1,2-distearyl-3-phosphate (13)



3-(2'-oleiyl glucosyl)-gallic acid (15)

Figure 1: Structural formulae of the compounds 9 - 15 isolated from the stem bark of Ficus religiosa The ¹H NMR spectrum of 15 exhibited two one-proton doublets at δ 7.43 (J = 2.9 Hz) and 7.27 (J = 2.9 Hz) assigned to *meta*-coupled aromatic H-2 and H-6, respectively. A one-proton doublet at δ 5.12 (J = 7.1 Hz) was attributed to anomeric H-1' proton. The other sugar proton signals appeared from δ 4.37 to 3.19. The vinylic protons of the fatty acid chain appeared as a two-proton multiplet at δ 5.35. A two-proton triplet at δ 2.38 (J = 7.2 Hz) was ascribed to methylene H₂-2" protons adjacent to the ester function. The other methylene protons appeared between δ 2.11 -1.24. A three-proton triplet at δ 0.94 (J = 6.3 Hz) was due to primary C-18" methyl protons. The ¹³C NMR spectrum



of **15** displayed signals for aromatic carbons between δ 160.06 - 114.07, ester carbon at δ 171.64 (C-1"), carboxylic carbon at δ 180.32 (C-7), anomeric carbon at δ 103.23 (C-1'), other sugar carbons from δ 77.63 to 61.73, vinylic carbons at δ 130.23 and 122.91 and methyl carbon at δ 14.17 (C-18"). The presence of the sugar H-2' proton in the ¹H NMR in the deshielded region at δ 4.13 and carbon C-2' signal at δ 74.36 suggested location of the ester linkage at C-2'. Acid hydrolysis of **15** yielded gallic acid, m. p. 258 – 260 °C, R_f 0.40 (toluene – ethyl acetate – formic acid – methanol, 3:3:0.8:0.2); D-glucose, R_f 0.26 (*n*-butanol- acetic acid - water, 4: 1: 5) and oleic acid, R_f 0.34 (85% glacial acetic acid). On the basis of these evidences, the structure of **15** has been elucidated as 2-(2'-octadec-9''-enoyl β -D-glucopyranosyl) gallic acid, a new gallic acid glucosidic oleate.

Conclusion

Phytochemical investigation of the stem bark of *Ficus racemosa* afforded glyceryl linolyldistearin (1), glyceryl 1linolyl-2-oleio-3-phosphate (2), glyceryl 1,2-dioleio-3-phosphate (3), gallic acid (4), (Z)-*n*-hexatriacont-12-en-1-ol (5), 6β -hydroxylupdienolyl 3 β -oleate (6), stigmast-5-en- 6α -ol-26-oic acid 3 β -linolenate (7) and glucuronosyl-($6\rightarrow 1'$)-glucoside (8). The stem bark of *Ficus religiosa* furnished arachidic acid (9), glyceryl-1-oleiyl-2-palmityl-3stearate (10), 1,2,3-trioleiyl β -D-arabinoside (11), glyceryl 1,2-diacyl -3-phosphates (12 - 14) and 2-(2'-oleiyl glucosyl) gallic acid (15). This work has enhanced understanding about the phytoconstituents of the stem barks of these *Ficus* species. These secondary metabolites can be used as analytical markers for quality control of the plants. Further research is recommended to screen bioactivities of the isolated phytoconstituents with a view for supplementing conventional drug development especially in developing countries.

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