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Isolation and structure elucidation of novel compounds from stem of *Dendrophthoe falcata*

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Dendrophthoe falcata (syn. With *Loranthus falcatus*) has been widely used in Indian and other traditional medicine systems as a therapeutic herb. Extensive pharmacological studies of leaf and stem extracts of *D. falcata* have shown significant biological activities. In the present study, an attempt has been made to carry out the phytochemical investigation of the petroleum ether and dichloromethane extract of the stem part of *D. falcata*. The isolation and identification of the possible bioactive phytochemicals of this plant may lead towards the development of new pharmaceutical products. Using repeated coloumn chromatography, preparative TLC and crystallization techniques, ten compounds have been isolated in pure form. On the basis of detailed spectral studies they have been identified as di-*iso*-octylphthalate **1**, nonadecan-1-ol **2**, stigmast-4-en-3-one **3**, stigmast-5-en-3β-ol **4**, stigmast-5,22-E-dien-3β-ol **5**, 3β-hydroxylup-20(29)-en-28-oic acid **6**, 3β-hydroxyolean-12-en-28-oic acid **7**, 7-hydroxy-4',5,6-trimethoxyflavone **8**, 2,3-dihydro-4-hydroxy-3,6,9-trimethylnaphtho[1,8-bc]pyran-7,8-dione **9** and 4-hydroxy-3-methoxybenzoic acid **10**. Compounds **1**, **4**, **5**, **7** and **10** have previously been reported from *Dendrophthoe species*. This is the first report of compounds **2**, **3**, **6**, **8** and **9** from *Dendrophthoe species* and the first report of **8** from a natural source. Further research work is needed on these identified compounds to explore their usefulness in phytopharmaceutical products.

Keywords: *Dendrophthoe falcata*, *Loranthus falcatus*, traditional medicinal use, plasticizer, flavonoid

Plants have long been a valuable source of biologically active natural products used for maintaining human health in traditional medicines. *Dendrophthoe falcata* (syn. With *Loranthus falcatus*), commonly known as the Indian mistletoe in English and *Banda* in Hindi, is an evergreen and shrubby, stem hemi-parasitic plant found on a variety of forest and fruit trees widely spread in India, Sri Lanka, Thailand, China and Australia[1,2]. It belongs to Loranthaceae family and is one of the seven species found in India[2]. It has been widely used in Ayurveda and other traditional medicine systems as a therapeutic herb in the treatment of pulmonary asthma, menstrual disorders, rheumatism, swellings, wounds and ulcers and is also known for its narcotic and diuretic properties[3].

Pharmacological studies of leaf and stem extracts of *D. falcata* have shown significant biological activities and also presence of phytoconstituents like steroids, flavonoids, glycosides, tannins and terpenoids[4,5]. Leaf extracts of *D. falcata* have shown *in vitro* antibacterial and antifungal activity[3], significant antihelminthic potency[6], anti-inflammatory activity especially for chronic inflammatory conditions such as rheumatoid arthritis[7], antifertility effect[8], inhibitory activity on α-amylase leading to antidiabetic effect[9], potential anti-carcinogenic properties and chemo-preventive effect[10]. The Indonesian mistletoe has been found to prevent proliferation which makes it a promising candidate in cancer therapeutics[11]. The high content of flavonoids and phenols found in leaf and stem extracts of *D. falcata* enable the same to be developed as natural antioxidants[5]. The ethanolic extract of its stems possesses anticonvulsant and muscle relaxant activity[12]. Some of the earlier[3-5,13] reported biologically important compounds isolated from *D. falcata* are shown in Figure 1.

To highlight the need for further investigation and better development of its medicinal properties, we have undertaken a detailed study of the chemical constituents of the stem part of *D. falcata*, collected from Assam, situated in the eastern part of India, with the purpose of isolating and identifying the possible bioactive phytochemicals for development of new pharmaceutical products.

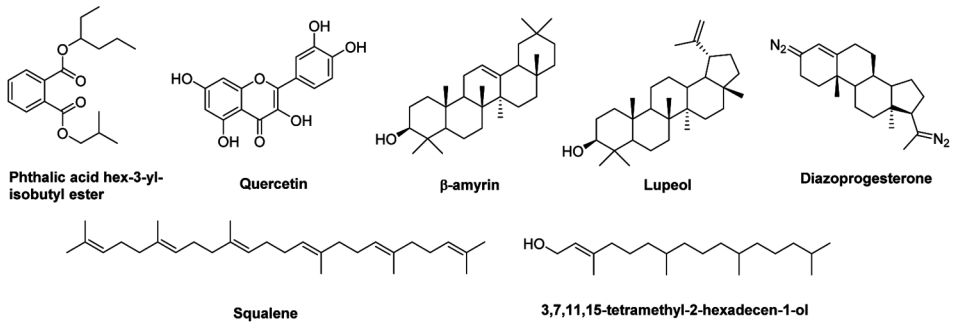

Figure 1 — Some earlier reported compounds from *Dendrophthoe falcata*

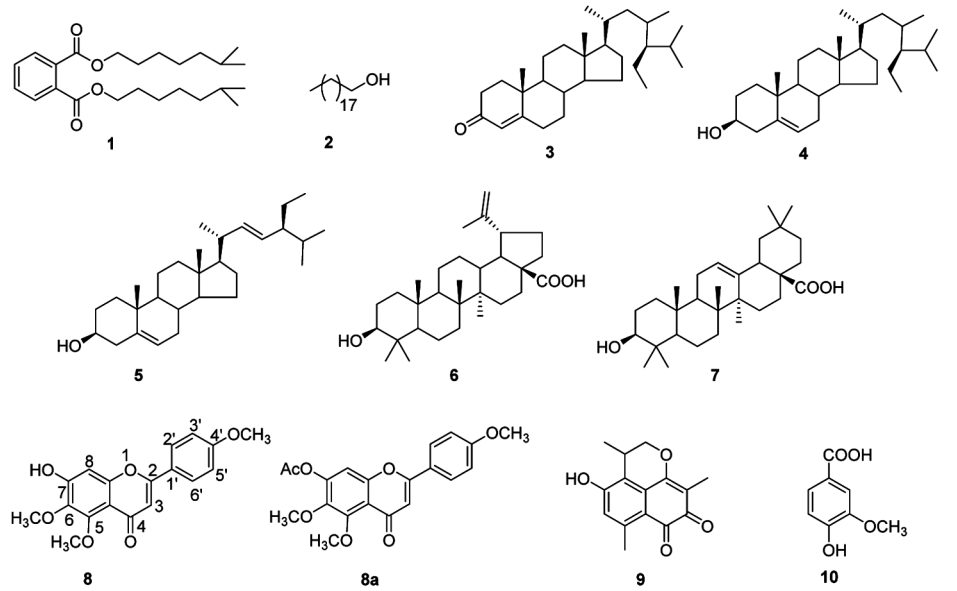

Figure 2 — Representative structures of compounds **1-10** isolated from *Dendrophthoe falcata*

Results and Discussion

Upon repeated column chromatography followed by crystallization, ten compounds **1-10** were isolated (Figure 2) from the combined petroleum ether and ethylacetate extract of stem part of *D. falcata*. Compound **1** was identified as 1,2-benzene dicarboxylic acid di-iso-octyl ester, also known as di-iso-octyl phthalate. Compound **1** has been reported earlier from the ethanol extract of leaves and barks of *D. falcata* and is known to have antimicrobial and antifouling activity[13]. Compound **4** was identified as stigmast-5-en-3β-ol or β-sitosterol[14]. Compound **5** was identified as stigmasta-5,22-*E*-dien-3β-ol or stigmasterol,[15] compound **7** was identified as 3β-hydroxy olean-12-en-28-oic acid or oleanolic acid[16] and compound **10** was identified as 4-hydroxy-3-methoxybenzoic acid or vanillic acid[17]. All the identifications were done on the basis of comparison of spectral data and melting point with that reported in literature.

Compound **2** was identified as nonadecan-1-ol[18] and compound **3** as stigmast-4-en-3-one or β-sitostenone[19], on the basis of comparison of spectral data and melting point with that reported in literature. This is the first report of nonadecan-1-ol and β-sitostenone from *Dendrophthoe Genus*. Earlier reports have shown the presence of unidentified alcohols in the leaf extracts of *D. falcata*[2]. Long chain primary alcohols including nona-1-decanol are reported to exhibit anti-bacterial activity[20]. β-sitostenone may be a necessary intermediate in the metabolism of β-sitosterol to other plant steroids in developing plants[21]. The mass spectrum of **6** was characteristic for a pentacyclic triterpene of the lupane series[22]. On the basis of comparison of spectral data and melting point with that reported in literature[23], **6** was identified

as 3β-hydroxy-lup-20(29)-en-28-oic acid or betulinic acid. This is the first report of occurrence of betulinic acid from *Dendrophthoe Genus* though the corresponding 3β-acetoxy-lup-20(29)-en-28-oic acid has been earlier reported[2].

Compound **8** was crystallized using petrol-acetone mixture as yellow needles. Its spot on the developed TLC plate (R_f= 0.60, chloroform/methanol 98:2) gave yellow color on spraying with sulphuric acid followed by heating, thus indicating a flavonoid structure. It also dissolved in conc. HCl, possibly due to the formation of an oxonium ion. In the UV-Vis spectrum, absorption maxima at 316 and 230 nm further suggested **8** to be a flavone. Absorption due to the C-4 carbonyl was observed at 1620 cm^{-1} in its IR spectrum. In the ^{1}H NMR spectrum, three singlets, each integrating for 3 protons, at δ 3.71, 3.86 and 3.88 along with a downfield signal at δ 10.09, integrating for one proton, showed the presence of three methoxy and one hydroxyl substituent. This was further confirmed by its acetylation followed by ^{1}H NMR and IR studies of the acetate **8a** obtained, which showed the presence of three $–OCH_3$ and one $–OCOCH_3$ groups. A significant downfield shift (δ 0.3-0.5) for the H-8 proton in **8a** indicated the position of a hydroxyl group at C-7 carbon of **8**. With a molecular ion peak at m/z 328, **8** was assigned the molecular formula $C_{18}H_{16}O_6$. The UV-Vis spectrum of **8** when recorded in the presence of aluminium chloride did not show any significant shift in either of the bands, thus indicating the absence of an adjacent dihydroxy system or a 3/5- hydroxyl group (chelated with C-4 carbonyl) in the molecule. A shift of 12 nm (230 to 242 nm) in band 2, in presence of sodium methoxide, indicated hydroxyl group at C-7 position and an insignificant shift of band 1 indicated the absence of a 4'-OH substituent. The 7-hydroxyl position was also supported by a bathochromic shift of band 2 in the presence of sodium acetate[24]. It's ^{1}H NMR spectrum displayed two characteristics doublets at δ 6.13 and 8.03,each for two protons, *J*=9.6 Hz and were assigned for H-3' & 5' and H-2' & 6' protons, respectively. A singlet at δ 6.54 was assigned for H-3 proton while that at δ 6.28 was assigned for H-8 proton. On the basis of above mentioned spectral data, **8** was suggested to be 7-hydroxy - 4',5,6-trimethoxyflavone. The relatively low maxima at 316 nm for band 1 observed in the UV-Vis spectrum also supported less substitution in B-ring. Mass fragmentation pattern of **8** also supported the proposed structure. This is the first report of isolation of 7-hydroxy-4',5,6-trimethoxyflavone from any natural source, though its corresponding 7-*O*-glycoside has been earlier reported from *Semecarpus kurzii*[25] and *Cladrastis shikokiana*,[26] which exhibit therapeutic effect on inflammatory bowel disease[27].

Compound **9** was crystallized from chloroform/ petrol to obtain its deep red shiny crystals. On basis of detailed spectral analysis, it was identified as a sequiterpenoid quinone, namely, mansonone H or 2,3-dihydro-4-hydroxy-3,6,9-trimethylnaphtho [11,8-bc] pyran-7,8-dione, reported earlier from *Mansonia attissima*[28] and roots of *Helicteres augustifolia*[29], a well-known tumor inhibitory plant found in Taiwan, and also from *Ulmus davidiana*[27] whose stem and root bark have been used as an oriental medicine for the treatment of edema, mastitis, gastric cancer and inflammation. Mansonone H is known to have anti- oxidative activity[30] and cytotoxic action by superoxide anion generation[31]. Occurrence of mansonone H in the stem of *D. falcata* makes it more important and useful for the treatment of gastric cancer and inflammation.

Experimental Section

The stem part (600g) of *Dendrophthoe falcata* was chipped, air dried and extracted for 28-30 hours with petroleum ether and dichloromethane, in succession, using a soxhlet apparatus. The solvent was removed from these extracts under reduced pressure and the residues examined separately on TLC using solvent systems of varying polarity and examined under UV light, in iodine atmosphere and by spraying with dilute sulphuric acid followed by heating. The petroleum ether and dichloromethane extracts were found to be similar in content and hence, were combined for further analysis.

The combined petroleum ether and dichloromethane extract (12g) was subjected to column chromatography. The column was prepared in petroleum ether using silica gel as an absorbent and eluted successively with petroleum ether, petroleum ether/ ethylacetate, ethylacetate and ethylacetate/methanol gradient in increasing polarity. A large number of fractions, each of 500 mL were collected and similar ones combined to make ten major fractions, the details of which are listed in Table I.

Characterization of 1,2-benzene dicarboxylic acid di-iso-octyl ester, 1

Compound **1** was obtained as colorless oil. R_f = 0.6 (5% ethyl acetate in petroleum ether). UV-Vis λ_{max}

Table I — Details of column chromatography eluent fractions

Fraction No.	Eluent	Nature of residue	TLC behaviour (Developing solvent)
I	Petroleum ether	Oil	Diffused spot (Petroleum ether)
II	Petroleum ether	pale yellow oil	1 alongwith minor impurities (Petroleum ether/ethylacetate, 95:5)
III	Petroleum ether	waxy white solid	Mainly 2 (Petroleum ether/ethylacetate, 90:10)
IV	Petroleum ether/ethylacetate (95:5)	dirty white solid	Mainly 3 and 4 (Petroleum ether/ethylacetate, 90:10)
V	Petroleum ether/ethylacetate (95:5) & (90:10)	white solid	Mainly5 (Petroleum ether /ethylacetate, 90:10)
VI	Petroleum ether/ethylacetate (90:10)	dirty white solid	Mainly6 and7 (Petroleum ether/ethylacetate,85:15)
VII	Petroleum ether/ethylacetate (85:15)	yellow solid	Mainly8 (Chloroform/methanol, 98:2)
VIII	Petroleum ether/ethylacetate (80:20)	deep red solid	Mainly9 (Chloroform/methanol, 90:10)
IX	Petroleum ether/ethylacetate (75:25)	white solid	Mainly 10 (Chloroform/methanol, 88:12)
X	Petroleum ether/ethylacetate (50:50) & (25:75)	brown resinous mass	

(MeOH): 274, 230 nm; IR (KBr): 2359, 1731, 1601, 1580, 1557, 1463, 1378, 1286, 1123, 1074, 1040, 965, 741, 704 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.70-0.91 (m, 30H, 2×-$(CH_2)_4CH(CH_3)_2$), 4.20 (m, 4H, 2×-αCH_2-), 7.43 (dd, 2H, *J* = 3.4 & 8.8 Hz, H-4 & H-5), 7.62 (dd, 2H, *J* = 3.4 & 8.8 Hz, H-3 & 6); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ 22.08, 25.04, 25.50, 27.41, 28.79, 35.34, 46.51, 66.07 (-OCH_2), 128.72 (C-4 & 5), 130.78 (C-3 & 6), 132.26 (C-1& 2), 167.55 (ArCHO); In the DEPT experiment, peaks at δ 25.04 and 25.50 were observed for methyl carbons while peaks in the region δ 27.41-66.07 were observed for methylene carbons. A peak at δ 22.08 was observed for a methine carbon; EI-MS: *m/z* (%) 390 (M^+, 2), 361 (2), 347 (2), 333 (2), 319 (2), 292 (2), 280 (10), 279 (47), 261 (5), 176 (2), 167 (17), 149 (100), 135 (2), 113 (27), 83 (8), 71 (28), 57 (30), 43 (15).

Characterization of nonadecan-1-ol, 2

Compound **2** was obtained as a white solid. m.p. 58-59°C (lit m.p. 60-61°C)[18]. R_f = 0.6 (10% ethyl acetate in petroleum ether). IR (KBr): 3442, 2923, 2855, 1466, 1375, 1360, 1059, 721, 666 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.89 (t, 3H, *J* = 6.9 Hz, -CH_3), 1.26-1.55 (brs, 32H, 16×-CH_2-), 1.56 (m, 2H, β-CH_2-), 3.65 (t, 2H, α-CH_2); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ 14.00, 22.59, 26.64, 29.61, 31.84, 32.74, 63.09; EI-MS: *m/z* (%) 284 (M^+, 9), 266 (M^+-H_2O, 6), 241 (16), 227 (18), 213 (6), 199 (16), 185 (26), 171 (16), 157 (14), 129 (42), 95 (42), 83 (63), 71 (100), 69 (42), 61 (88), 57 (66), 55 (76).

Characterization of stigmast-4-en-3-one, 3

Compound **3** was obtained as colorless needless. m.p. 86-87°C (lit m.p. 88°C) [19]. R_f = 0.8 (10% ethyl acetate in petroleum ether). UV-Vis λ_{max} (MeOH): 241 nm; IR (KBr): 2928, 2851, 1679, 1618, 1464, 1377, 1301, 1276, 720 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.71 (s, 3H, 18-CH_3), 0.82 (d, 6H,*J* = 6.5 Hz, 26 & 27-CH_3), 0.84 (t, 3H, *J* = 6.7 Hz, 29-CH_3), 0.91 (d, 3H, *J* = 6.4 Hz, 21-CH_3), 1.18 (s, 3H, 19-CH_3), 1.50-2.35 (m, 27H), 2.38 (m, 2H, -$COCH_2$-), 5.72 (s, 1H, H-4); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ 10.86 (C-29), 16.29 (C-19), 17.93 (C-6), 18.70 (C-21), 19.94 (C-18), 21.99 (C-11), 23.09 (C-26 & 27), 25.03 (C-23), 27.09 (C-15), 28.08 (C-25), 28.60 (C-12), 30.97 (C-28), 31.86 (C-2), 32.81 (C-7), 32.88 (C-8), 34.55 (C-20), 34.60 (C-22), 35.02 (C-10), 38.54 (C-1), 39.69 (C-24), 40.01 (C-16), 44.76 (C-13), 52.74 (C-9), 54.80 (C-17), 54,94 (C-14), 114.14 (C-4), 122.64 (C-5), 169.86 (C-3); EI-MS *m/z* (%): 412 (M^+, 100), 397 (12), 394 (4), 355 (8), 201 (6), 167 (10), 175 (10), 147 (20), 135 (18), 124 (92), 107 (17), 95 (22), 81 (15), 55 (20), 43 (22).

Characterization of stigmast-5-en-3β-ol, 4

Compound **4** was obtained as colourless needless. m.p. 134°C (lit m.p. 136-137°C) [14]. R_f = 0.50 (10% ethyl acetate in petroleum ether). IR (KBr): 3550, 3090, 2965, 1627, 1470, 1370, 1055, 1025, 930, 927, 880, 803 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.68 (s, 3H, 18-CH_3), 0.81 (d, 6H,*J* = 6.8 Hz, 26 & 27-CH_3), 0.84 (t, 3H, *J* = 7.0 Hz, 29-CH_3), 0.89 (d, 3H, *J* = 6.3

Hz, 21-CH_3), 0.99 (s, 3H, 19-CH_3), 1.34-2.32 (m, 29H), 3.22 (m, 1H, H-3α), 5.38 (t, 1H, *J* = 6.9 Hz, H-6); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ 11.31 (C-29), 12.07 (C-19), 18.88 (C-21), 19.45 (C-18), 21.21 (C-11), 22.50 & 22.82 (C-26 & 27), 24.15 (C-23), 24.34 (C-15), 28.08 (C-25), 28.39 (C-12), 29.31 (C-28), 31.61 (C-2), 32.02 (C-7), 32.33 (C-8), 35.82 (C-20), 36.46 (C-22), 36.57 (C-10), 37.51 (C-1), 39.69 (C-24), 40.01 (C-16), 44.44 (C-4 &13), 50.58 (C-9), 56.51 (C-17), 71.33 (C-3), 121.36 (C-6), 141.26(C-5); EI-MS: *m/z* (%) 414 (M^+, 57), 399 (M^+-CH_3, 12), 396 (M^+-H_2O, 12), 381 (M^+-H_2O-CH_3, 6), 329 (9), 303 (14), 273 (M^+-$C_{10}H_{21}$, 19), 255 (12), 232 (M^+-$C_{10}H_{21}$-ring D, 16), 213 (12), 173 (8), 164 (223-ring C-19CH_3, 18), 145 (20), 133 (19), 107 (35), 95 (37), 81 (45), 70 (50), 57 (50), 55 (70), 43 (100), 41 (30).

Characterization of stigmasta-5,22-*E*-dien-3β-ol, 5

Compound **5** was obtained as colorless needles. m.p. 167-168°C (lit m.p. 170°C) [15]. R_f = 0.30 (10% ethyl acetate in petroleum ether). IR (KBr): 3532, 3095, 2960, 1679, 1618, 1462, 1367, 1053, 1031, 970, 925, 872, 822 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ 0.66 (s, 3H, 18-CH_3), 0.78 (d, 6H, *J* = 6.8 Hz, 27-CH_3), 0.79 (t, 3H, *J* = 7.2 Hz, 29-CH_3), 0.80 (s, 3H,19-CH_3), 0.84 (d, 3H, *J* = 6.8 Hz, 26-CH_3), 1.00 (d, 3H, *J* = 6.5 Hz, 21-CH_3), 3.22 (m, 1H, H-3α), 5.31 (m, 3H, olefinic-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ 12.02 (C-29), 12.21 (C-18), 19.01 (C-26), 19.43 (C-19), 21.16 (C-11, 21 & 27), 24.47 (C-15), 25.45 (C-28), 28.91 (C-16), 31.70 (C-2), 31.92 (C-7, 8 & 25), 36.61 (C-10), 37.48 (C-1), 39.81 (C-12), 40.50 (C-20), 42.43 (C-4 & 13), 50.39 (C-9), 51.32 (C-24), 56.07 (C-17), 57.00 (C-14), 71.87 (C-3), 121.72 (C-6), 129.43 (C-23), 138.44 (C-22), 140.91 (C-5); EI-MS: *m/z* (%) 412 (M^+, 55), 397 (M^+-CH_3, 10), 394 (M^+-H_2O, 11), 379 (M^+-H_2O-CH_3, 5), 314 (M^+-C_7H_{13}-H, 20), 285 (M^+-C_9H_{17}-2H, 22), 232 (M^+-C_9H_{17}-ring D, 18), 213 (13), 164 (223-ring C-19CH_3, 20),145 (20), 133 (19), 107 (35), 95 (37), 81 (45), 70 (50), 57 (50), 55 (70), 43 (100), 41 (30).

Characterization of 3β-hydroxy-lup-20(29)-en-28-oic acid, 6

Compound **6** was crystallized from chloroform/ methanol as colorless needles, lit m.p. 277-278°C[23]. R_f = 0.70 (15% ethyl acetate in petroleum ether). IR (KBr): 3500, 3000, 2925, 1698, 1650, 1460, 1382, 1326, 1250, 1184, 1103, 1031, 982, 890 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ 0.75, 0.82, 0.93, 0.97 and 1.25 (s, each, 15H, 5×-CH_3), 1.68 (s, 3H, vinylic-CH_3), 3.18 (dd, 1H, *J* = 5.3 & 10.8 Hz, H-3α), 4.61 & 4.73 (bts, 1H each, =CH_2); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ 14.58 (C-27), 15.23 (C-24), 15.92 (C-25), 16.02 (C-26), 18.18 (C-6), 19.27 (C-30), 20.75 (C-11), 25.40 (C-12), 27.32 (C-2), 27.90 (C-23), 29.62 (C-21), 30.46 (C-15), 32.06 (C-16), 34.25 (C-7), 36.95 (C-22), 37.14 (C-10), 38.31 (C-13), 38.65 (C-1), 38.80 (C-4), 40.63 (C-8), 42.38 (C-14), 46.82 (C-18), 49.22 (C-19), 50.47 (C-9), 55.31 (C-5), 56.24 (C-17), 79.01 (C-3), 109.76 (C-29), 150.52 (C-20), 180.15 (C-28); EI-MS: *m/z* (%) 456 (M^+, 40), 441 (M^+-CH_3, 5), 438 (M^+-H_2O, 27), 423 (M^+-H_2O-CH_3, 10), 411 (M^+-45, 18), 395 (10), 369 (5), 302(7), 262 (12), 248 (52), 220 (28), 219 (20), 207 (52), 203 (32), 189 (100), 175 (26), 135 (36), 121 (23), 95 (28), 69 (15).

Characterization of 3β-hydroxyolean-12-en-28-oic acid, 7

Compound **7** was crystallized from chloroform/ methanol as colorless needles, lit m.p. 310°C[16]. R_f = 0.50 (15% ethyl acetate in petroleum ether). IR (KBr): 3420, 2900, 2840, 1701, 1464, 1390, 1366, 1347, 1325, 1305, 1264, 828, 818, 804 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ 0.75 (s, 3H, 26-CH_3), 0.80 (s, 6H, 23 & 24-CH_3), 0.92 (s, 6H,29 & 30- CH_3), 1.00 (s, 3H,27-CH_3), 3.20 (m, 1H, H-3α), 5.27 (m, 1H, H-12); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ 15.31 (C-25), 15.64 (C-24), 16.86 (C-26), 18.31 (C-6), 23.10 (C-11), 23.49 (C-16), 23.61 (C-30), 26.02 (C-27), 27.73 (C-15), 27.44 (C-2), 28.10 (C-23), 30.65 (C-20), 32.31 (C-22), 32.62 (C-7), 33.14 (C-29), 33.80 (C-21), 37.03 (C-10), 38.58 (C-1), 38.73 (C-4), 39.33 (C-8), 41.39 (C-18), 41.60 (C-14), 45.87 (C-19), 46.68 (C-17), 47.58 (C-9), 55.29 (C-5), 78.74 (C-3), 122.10 (C-12), 143.44 (C-13), 181.02 (C-28); EI-MS: *m/z* (%) 456 (M^+, 54), 441 (M^+-CH_3, 11), 411 (M^+-3×CH_3, 12), 395 (12), 300 (7), 248 (45), 207 (35), 203 (100), 189 (41), 175 (8), 134 (19), 107 (32), 95 (26), 81 (45), 70 (45), 57 (52), 55 (60), 43 (60), 41 (30).

Characterization of 7-hydroxy-4',5,6-trimethoxyflavone, 8

Compound **8** was crystallized from petrol/acetone as yellow needles, m.p. 205-206°C. R_f = 0.60 (2% methanol/chloroform). UV-Vis λ_{max} (MeOH): 316, 230 nm; IR (KBr): 3321, 2928, 1704, 1620, 1563, 1506, 1464, 1359, 1268, 1201, 1142, 1118, 1022, 928, 817 cm^{-1}; ^{1}H NMR (300 MHz, Acetone-d_6): δ 3.71, 3.86 and 3.88 (s, each, 9H, 3×-OCH_3), 6.13 (d, 2H, *J* = 9.6 Hz, H-3' & 5'), 6.28 (s, 1H, H-8), 6.54 (s, 1H,

H-3), 8.02 (d, 2H, J = 9.6 Hz, H-2' & 6'), 10.09 (s, 1H, -OH); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ 55.38, 55.48, 59.83 (3×-OCH_3), 96.91 (C-8), 106.42 (C-3), 109.77 (C-3' & 5'), 18.18 (C-6), 124.45 (C-1'), 127.82 (C-6), 138.68 (C-2' & 6'), 144.63 (C-5), 148.09 (C-7), 155.81 (C-2), 157.41 (C-4'), 190.02 (C-4); EI-MS: *m/z* (%) 328 (M^+, 90), 313 (25), 300 (17), 197 (42), 185 (21), 168 (38), 135 (47), 132 (41), 107 (65), 95 (42), 81 (38), 69 (32).

Acetylation of compound 8

Compound **8** (20 mg) was dissolved in dry pyridine (2 mL) in a round bottom flask and dry acetic anhydride was added drop-wise. The reaction mixture was heated over a bath for few minutes and then kept at RT for overnight. After completion, the product formed was crystallized using DCM/MeOH to get colorless needles, m.p. 197-199°C. R_f = 0.40 (chloroform). IR (KBr): 2935, 1761, 1710, 1624, 1575, 1492, 1412, 1360, 1280, 1218, 1195, 1136, 1120, 1025, 919, 823 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ 2.37 (s, 3H, -$OCOCH_3$) 3.88, 3.89 and 3.99 (s, each, 9H, 3×-OCH_3), 6.22 (d, 2H, J = 9.6 Hz, H-3' & 5'), 6.45 (s, 1H, H-3), 6.65 (s, 1H, H-8), 7.96 (d, 2H, J = 9.6 Hz, H-2' & 6'). The above mentioned spectral data of the acetate derivative of **8** was in accordance with that expected for 7-acetoxy-4',5,6-trimethoxy flavone.

Characterization of 2,3-dihydro-4-hydroxy-3,6,9-trimethylnaphtho[11,8-bc]pyran-7,8-dione, 9

Compound **9** was crystallized from petrol/ chloroform as deep red shiny crystals, lit m.p. 356°C[29]. R_f = 0.50 (10% methanol/chloroform). UV-Vis λ_{max} (MeOH): 404, 297, 209 nm; IR (KBr): 3263, 2925, 1670, 1634, 1612, 1583, 1457, 1426, 1379, 1353, 1320, 1267, 1227, 1186, 1157, 1116, 1056, 1030, 949, 892 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 1.24 (d, 3H, J = 6.8 Hz, 3-CH_3), 1.83 (s, 3H, 9-CH_3), 2.51 (s, 3H, 6-CH_3), 3.20 (m, 1H, H-3), 4.28 (dd, 1H, J = 3.1 & 10.8 Hz, H-2a), 4.41 (brs, 1H, J = 10.8 Hz, H-2b), 6.75 (s, 1H, H-5), 11.17 (brs, 1H, 4-OH); ^{13}C NMR (75.5 MHz, DMSO-d_6): δ 7.11 (9-CH_3), 16.26 (3-CH_3), 22.02 (6-CH_3), 24.95 (C-3), 70.99 (C-2), 113.98 (C-9), 118.43 (C-5), 143.94 (C-6), 159.59 (C-4), 179.69 & 180.22 (C-7 & 8); EI-MS: *m/z* (%) 260 (M^++2, 82), 258 (M^+, 20), 245 (45), 230 (100), 215 (40), 201 (18), 187 (13), 159 (12), 158 (12), 129 (12), 116 (18), 91 (6), 83 (6), 77 (6), 55 (10), 44 (40).

Characterization of 4-hydroxy-3-methoxybenzoic acid, 10

Compound **10** was crystallized as colorless needles, m.p. 208°C (lit m.p. 210°C) [17]. R_f = 0.40 (12% methanol/chloroform); IR (KBr): 3500, 2998, 1680, 1600, 1488, 1307, 1244, 1223, 1190, 1101, 1021, 899, 877, 798, 755 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 3.86 (s, 3H, -OCH_3), 6.84 (d, 1H, J = 8.6 Hz, H-5), 7.46-7.49 (m, 2H, H-2 & 6), 9.49 (brs, 1H, -OH); ^{13}C NMR (75.5 MHz, DMSO-d_6): δ 56.34 (-OCH_3), 113.51 (C-5), 115.61 (C-6), 124.29 (C-2), 126.12 (C-1), 151.78 (C-4), 152.01 (C-3), 168.15 (-COOH); EI-MS: *m/z* (%) 168 (M^+, 100), 153 (M^+-CH_3,46), 151 (21), 125 (12), 123 (M^+-COOH, 15), 108 (M^+-COOH-CH_3, 13), 97 (23), 84 (10), 79 (8), 77 (5), 66 (10), 52(9).

Conclusions

Ten compounds **1-10** were isolated in pure form from the combined petroleum ether and dichloromethane extract of stem part of *Dendrophthoe falcata* and all compounds were characterized on basis of their spectral analysis. These may be categorized as one plasticizer (**1**), one long chain alcohol (**2**), three triterpenoids (**3**, **4**, **5**), one pentacyclic triterpene of lupane series (**6**), one triterpenoid with oleanene skeleton (**7**), one flavonoid (**8**), one sesquiterpenoid quinone (**9**) and one phenolic compound (**10**). Compounds **1**,**4**,**5**,**7** and **10** have been reported earlier from *Dendrophthoe falcata.* This is the first report of **2**,**3**,**6**,**8** and **9** from *Dendrophtho esp.* This is also the first report of **8** from any natural source though its 7-*O*-glycoside has earlier been reported. The occurrence of such physiologically active substances in *Dendrophthoe falcata* not only justify its use in Ayurvedic system of medicine but also suggests the need for further research on these compounds to explore their potential as phyto pharmaceutical products.

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