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Phenyl alkyls and citronellyl glucosides from the silk cocoons of *Bombyx mori* L.

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ABSTRACT

*Silk cocoons, produced by Bombyx mori L. (Bombicidae) are used to treat heart palpitation, cough, asthma and catarrh. Phytochemical investigation of the cocoons led to the isolation of alkyl substituted dioxymethylene benzene derivatives and two citronellyl glucosides for the first time characterized as n-pentanyl-3,4-(dioxymethylene) benzene (**1**), n-pentanyl-2,3-dioxymethylene-4-hydroxybenzene (**2**), n-decanyl-3,4-dioxymethylene-4-hydroxybenzene (**3**), 3,6,10-trimethyldodecan-1-ol-1β-D-glucopyranoside (**4**) and 8-hydroxydihydrocitronelloyl-1β-D-glucopyranosyl-(2′→1″)-β-D-glucuronopyranosyl-2″-(3‴-hydroxy-4‴-acetoxy)benzoate (**5**) on the basis of spectral data analysis.*

Keywords: *Bombyx mori*, silk cocoons, alkyl dioxymethylene benzene, citronellyl glucosides.

INTRODUCTION

Silk cocoons, known as abresham, are produced by a domesticated monophagous insect silk moth, *Bombyx mori* L. (Bombicidae) whose only food is mulberry leaves. The transparent and waxy larva spins its silk cocoon and moves about its head in all directions instinctively feeling for some niche to which the exuding silk fibre can be safely attached [1]. The cocoon shell consists of silk fibroin fibre (70 %) surrounded by a sericin layer made up of sericin (25 %) and nonprotein compounds. The non-sericin component is consisted of carbohydrate, wax, flavonoids and pigments [2-4]. Flavanoids have been found as pigments in the cocoon shells of some silk worm races [5-7]. Larvae of the silk worm sequester flavonoids into their cocoons from the leaves of their host plant, *Morus alba*, family Moraceae [5]. However, flavonol glycosides were not present in the mulberry leaves, but isolated from the cocoons [7]. Therefore, it was inferred that flavonoids absorbed from their diet are modified within the insects by a glucosyl transferase that can transfer a glucose residue to the C-5 hydroxy position of quercetin for using these compounds to increase fitness and to help increase the antioxidative state of tissue [8]. An unsaturated fat with a high concentration of phospholipid was present in the silkworm [9]. In insects the formation of glycosides is the predominant pathway for dietary flavonoids [10,11] and glycosylation of polyphenolics in insects is catalyzed by UDP-glucosyl-transferase (UGT) [12,13] suggesting the possibility that a UGT enzyme that can transfer a glucose moiety to the C-5 position of the flavonoid is functioning in *B. mori*. A correlation between the quantitative changes in L-methionine analogs, the ratio of D-serine and L-serine during the pupal stage, and metamorphosis was observed in the pupae of *B. mori*. [14]. The alkaloid constituents 1-deoxynojirimycine, fagomine and 3-epifagomine were isolated from silkworm dropping [15]. In traditional medicine, ash of the silkworm and cocoon is used as aphrodisiac and rejuvenating tonic [16]. The silk cocoons are useful as hypotensive, expectorant, bronchodilator and attenuant drug and to treat palpitation of heart, cough, asthma and catarrh [17]. This manuscript describes isolation and characterization of phenolic compounds from the silk cocoons produced by *Bombyx mori*.

MATERIALS AND METHODS

General

The melting points were determined on a Perfit apparatus and are uncorrected. The IR spectra were recorded on KBr pellet using a Jasco FT/IR-5000 instrument (FTS 135, Hongkong). The ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz) spectra were scanned on Avance DRX 400, Bruker spectrospin 400 MHz instrument (Rheinstetten, Germany) using $CDCl_3$ as solvent and TMS as internal standard. FAB-MS were measured using JEOL-JMS-DX 303 spectrometer (Peabody, MA, USA). Column (450×4×0.2 cm) chromatography was performed on silica gel (60-120 mesh, Qualigens, Mumbai, India) and thin layer chromatography on silica gel G-coated TLC plates (Merck). Spots were visualized by exposure to iodine vapours, UV radiation and by spraying with ceric sulphate solution.

Plant material

The silk cocoons of *Bombyx mori* were procurred from local market of Delhi, Khari Baoli and authenticated by Dr. Parvez Akhtar, Taxonomist, Central Council for Research in Unani Medicine, Jamia Hamdard (Hamdard University). A voucher specimen No. PRL/JH/08/49 is deposited in the herbarium of the Phytochemistry Research Laboratory, Jamia Hamdard, New Delhi, India.

Extraction and isolation

The cocoons of *B. mori* were opened to remove the dead pupae. The cocoon shell were dried at 45 ºC for 3 days and coarsely powdered. The powdered cocoon shells (3 kg) were extracted exhaustively with ethanol (95 %) in a Soxhlet apparatus. The ethanolic extract was concentrated under reduced pressure to yield dark yellow, viscous syrupy mass (150 g, 5.0 %). It was dissolved in minimum amount of methanol and adsorbed on silica gel (60-120 mesh) for preparation of slurry. The slurry was dried in air and chromatographed over silica gel column packed in petroleum ether. The column was eluted with petroleum ether, petroleum ether-chloroform (9:1, 3:1, 1:1, and 1:3 *v/v*), chloroform and chloroform-methanol (99:1, 49:1, 19:5, 9:1, 3:1, 1:1 *v/v*) in order of increasing polarity to isolate the following compounds:

n- Pentanyl-3,4-(dioxymethylene) benzene (1)

Elution of the column with chloroform-methanol (99:1) furnished colourless crystals of **1**, recrystallized with chloroform:methanol (1:1), 103 mg (0.003 % yield), R_f: 0.68 (chloroform:methanol, 9:1); m.p. 174-175 ºC; UV λ_{max} (MeOH): 271 nm (log ε 4.8); IR ν_{max} (KBr): 2955, 2845, 1590, 960, 791 cm^{-1}; ^{1}H NMR (DMSO-d_6): δ 8.29 (1H, d, *J*=9.5 Hz, H-5), 8.08 (1H, d, *J*=2.5 Hz, H-2), 7.97 (1H, m, H-6), 3.39 (2H, br, O-CH2-O), 2.54 (2H, t, J=7.2 Hz, H_2-7), 1.98 (2H, m, H_2-8), 1.48 (2H, m, H2-9), 1.21 (2H, brs, H_2-10), 0.83 (3H, t, J=6.5 Hz, Me-11); ^{13}C NMR (DMSO-d_6): δ 148.91 (C-1), 144.37 (C-2), 155.16 (C-3), 151.16 (C-4), 141.77 (C-5), 138.10 (C-6), 31.63 (C-7), 28.71 (C-8), 28.71 (C-9), 24.11 (C-10), 14.36 (C-11), 100.05 (-O-CH2O-); +ve FAB MS *m/z* (rel. int): 181 $[M+H]^+$ ($C_{12}H_{17}O_2$) (11.2).

n-Pentanyl-2,3-dioxymethylene-4-hydroxybenzene (2)

Elution of the column with chloroform-methanol (19:1) gave colourless crystals of **2**, recrystallized with chloroform:methanol (1:1), 256 mg (0.007 % yield), R_f: 0.65 (chloroform:methanol, 4:1); m.p. 188-189 ºC; UV λ_{max} (MeOH): 276 nm (log ε 5.1); IR ν_{max} (KBr): 3490, 2952, 2843, 1556, 1411, 1228, 1025, 963, 854 cm^{-1}; ^{1}H NMR (DMSO-d_6): δ 7.35 (1H, d, *J*=7.8 Hz, H-5), 5.46 (1H, d, *J*=7.8 Hz, H-6), 3.39 (2H, brs, O-CH_2-O), 2.51 (2H, t, *J*=7.2 Hz, H_2-7), 1.22 (6H, brs, 3×CH_2), 0.83 (3H, t, *J*=6.1 Hz, Me-11); ^{13}C NMR (DMSO-d_6): δ 144.37 (C-1), 155.27 (C-2), 151.29 (C-3), 164.06 (C-4), 141.77 (C-5), 140.13 (C-6), 31.73 (C-7), 28.68 (C-8, C-9), 23.96 (C-10), 14.26 (C-11), 100.05 (-O-CH2O-); +ve FAB MS *m/z* (rel. int): 209 $[M+H]^+$ ($C_{12}H_{17}O_3$).

n-Decanyl-3,4-dioxymethylene-4-hydroxybenzene (3)

Further elution of the column with chloroform-methanol (19:1) afforded colourless crystals of **3**, recrystallized from chloroform-methanol (1:1), 159 mg (0.004 % yield), R_f: 0.60 (chloroform:methanol, 4:1); m.p. 210-212 ºC; UV λ_{max} (MeOH): 275 nm (log ε 4.2); IR ν_{max} (KBr): 3398, 2950, 2845, 1541, 1526, 1460, 1250, 930 cm^{-1}; ^{1}H NMR (DMSO-d_6): δ 7.37 (1H, d, *J*=7.9 Hz, H-5), 5.47 (1H, d, *J*=7.9 Hz, H-6), 3.37 (2H, brs, O-CH_2-O), 2.41 (2H, t, *J*=7.1 Hz, H_2-7), 1.84 (2H, m, H_2-8), 1.22 (14H, brs, 7×CH_2), 0.85 (3H, t, *J*=6.1 Hz, Me-16); ^{13}C NMR (DMSO-d_6): δ 142.38 (C-1), 151.72 (C-2), 151.72 (C-3), 164.59 (C-4), 140.73 (C-5), 137.91 (C-6), 33.81 (C-7), 31.49 (C-8), 29.16 (C-9), 29.16 (C-10), 29.16 (C-11), 29.16 (C-12), 26.73 (C-13), 24.62 (C-14), 22.45 (C-15), 14.26 (C-16), 100.39 (O-CH_2O-); +ve FAB MS *m/z* (rel. int): 279 $[M+H]^+$ ($C_{17}H_{27}O_3$) (2.3), 137 (74.9).

Dihydrocitronellyl glucoside (4)

Elution of the column with chloroform-methanol (9:1) yielded colourless crystals of **4**, recrystallized from chloroform-acetone (1:1), 250 mg (0.0069 % yield), R_f: 0.80 (chloroform:methanol, 1:1); m.p. 298-299 ºC; UV λ_{max} (MeOH): 224 nm (log ε 4.9); IR ν_{max} (KBr): 3450, 3285, 2955, 2845, 1470, 1210, 1116 cm^{-1}; ^{1}H NMR (DMSO-d_6): δ

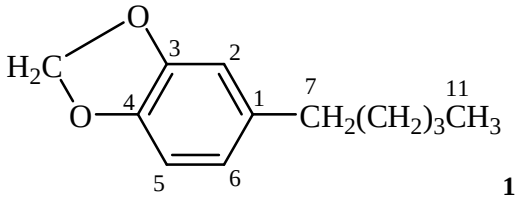
5.12 (1H, d, *J*=7.2 Hz, H-1′), 4.25 (1H, m, H-5′), 3.98 (1H, m, H-2′), 3.56 (1H, m, H-3′), 3.43 (1H, m, H-4′), 3.39 (1H, d, *J*=8.1 Hz, H_2-1a), 3.35 (1H, d, *J*=6.3 Hz, H_2-1b), 3.13 (1H, d, *J*=6.1 Hz, H_2-6′a), 3.10 (1H, d, *J*=6.1 Hz, H_2-6′b), 2.50 (2H, m, H_2-2), 2.26 (1H, m, H-6), 2.09 (1H, m, H-10), 1.82 (2H, m, H_2-3), 1.69 (2H, m, H_2-5), 1.59 (2H, m, H_2-7), 1.44 (2H, m, H_2-8), 1.29 (6H, brs, H_2-4, H_2-9, H_2-11), 1.23 (3H, d, *J*=6.3 Hz, Me-13), 0.96 (3H, d, *J*=6.0 Hz, Me-14), 0.92 (3H, d, *J*=6.7 Hz, Me-15), 0.83 (3H, t, *J*=6.3 Hz, Me-12); ^{13}C NMR (DMSO-d_6): *δ* 65.51 (C-1), 58.14 (C-2), 53.11 (C-3), 52.09 (C-4), 28.58 (C-5), 54.96 (C-6), 31.36 (C-7), 30.88 (C-8), 28.56 (C-9), 54.32 (C-10), 28.59 (C-11), 13.34 (C-12), 28.23 (C-13), 24.98 (C-14), 21.62 (C-15), 103.53 (C-1′), 73.58 (C-2′), 69.98 (C-3′), 68.21 (C-4′), 78.70 (C-5′), 63.46 (C-6′); +ve FAB MS *m/z* (rel. int): 381 $[M+H]^+$ ($C_{21}H_{33}O_6$) (2.3), 217 (5.6), 200 (6.5).

Dihydrocitronelloyl glucuronosidic ester (5)

Elution of the column with chloroform-methanol (9:1) gave pale yellow crystals of **5**, recrystallized from chloroform-methanol (1:1), 153 mg (0.0039 % yield), R_f: 0.65 (chloroform:acetone, 3:2); m.p. 278-279 ºC; UV λ_{max} (MeOH): 225 nm (log ε 5.3); IR ν_{max} (KBr): 3450, 3310, 2955, 2850, 1735, 1721, 1690, 1560, 1390, 1210, 1172, 950 cm^{-1}; ^{1}H NMR (DMSO-d_6): *δ* 7.79 (1H, d, *J*=2.5 Hz, H-2‴), 7.71 (1H, d, *J*=8.9 Hz, H-5‴), 7.58 (1H, dd, *J*=2.5, 8.9 Hz, H-6‴), 5.32 (1H, d, *J*=7.1 Hz, H-1′), 5.21 (1H, d, *J*=7.3 Hz, H-1″), 4.23 (1H, m, H-2″), 4.17 (1H, m, H-2′), 4.03 (1H, m, H-3′), 3.98 (1H, m, H-3″), 3.91 (2H, m, H-4′, H-4″), 3.13 (2H, d, *J*=6.5 Hz, H_2-8), 2.50 (3H, brs, $OCOCH_3$), 2.26 (2H, d, J=7.1 Hz, H_2-2), 2.05 (2H, m, H_2-4), 1.95 (1H, m, H-3), 1.69 (2H, m, H_2-5), 1.62 (1H, m, H-7), 1.56 (2H, m, H_2-6), 1.23 (3H, d, *J*=6.5 Hz, Me-9), 0.86 (3H, d, *J*=6.1 Hz, Me-10); ^{13}C NMR (DMSO-d_6): *δ* 175.42 (C-1), 56.48 (C-2), 53.41 (C-3), 37.28 (C-4), 31.32 (C-5), 29.20 (C-6), 49.96 (C-7), 60.78 (C-8), 25.31 (C-9), 17.13 (C-10), 105.23 (C-1′), 71.53 (C-2′), 69.91 (C-3′), 66.90 (C-4′), 74.02 (C-5′), 179.43 (C-6′), 101.6 (C-1″), 79.36 (C-2″), 68.90 (C-3″), 64.07 (C-4″), 73.71 (C-5″), 177.20 (C-6″), 130.47 (C-1‴), 124.89 (C-2‴), 157.76 (C-3‴), 156.98 (C-4‴), 146.13 (C-5‴), 115.39 (C-6‴), 173.77 (C-7‴), 171.92 ($OCOCH_3$); +ve FAB MS *m/z* (rel. int): 719 $[M+H]^+$ ($C_{31}H_{43}O_{19}$) (1.6), 523 (6.8), 187 (14.2), 179 (18.1), 171 (25.3), 155 (61.9).

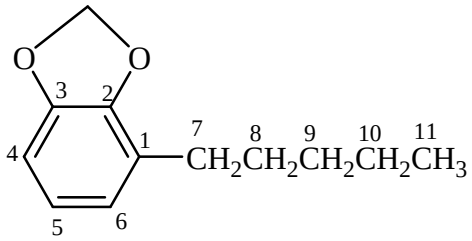
RESULTS AND DISCUSSION

Compound **1**, named *n*-pentanyl-3,4-(methylene dioxy)benzene, was obtained as a colourless crystalline mass from choloroform-methanol (99:1) eluents. It had a UV absorption maximum at 271 nm and IR absorption bands at 1590 and 960 cm^{-1} indicating aromatic nature of the compound. On the basis of FAB mass and ^{13}C NMR spectra the molecular ion peak of **1** was determined at *m/z* 181 [M+H]+ ($C_{12}H_{17}O_2$). The ^{1}H NMR spectrum of **1** showed two one-proton doublets at *δ* 8.29 (*J*=9.5 Hz) and 8.08 (*J*=2.5 Hz) assigned to aromatic *ortho*-coupled H-5 and meta-coupled H-2 and a one-proton multiplet at *δ* 7.97 due to H-6. A two-proton broad signal at *δ* 3.39 and a two-proton triplet at *δ* 2.54 (*J*=7.2 Hz) were ascribed to dioxomethylene and methylene H_2-7 protons linked to the aromatic ring, respectively. Three two-proton signals at *δ* 1.98 (m), 1.48 (m) and 1.21 (brs) were attributed to the remaining methylene protons. A three-proton triplet at *δ* 0.83 (*J*=6.5 Hz) was accounted to C-11 primary methyl protons. The ^{13}C NMR spectrum of **1** exhibited signals for aromatic carbons between *δ* 155.16-138.10, methylene carbons from *δ* 31.63 to 24.11, dioxo-methylene carbon at *δ* 100.05 and methyl carbon at *δ* 14.36 (C-11). On the basis of spectral data analysis the structure of **1** has been characterized as *n*- pentanyl-3,4-(dioxymethylene) benzene. This is a new dioxomethylene benzene derivative.



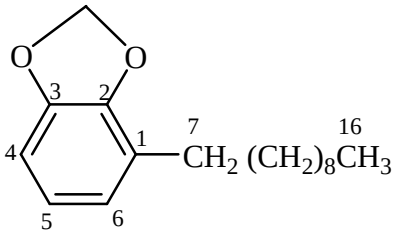
1

Compound **2**, named *n*-pentanyl-3,4-(methylenedioxy)phenol, was obtained as a colourless crystalline mass from chloroform-methanol (19:1) eluents. It gave positive tests for phenols and had UV absorption maximum at 276 nm and IR absorption bands at 3490 cm^{-1} for hydroxyl group and at 1556 and 963 cm^{-1} for aromatic ring. Its molecular ion peak was determined at *m/z* 209 $[M+H]^+$ on the basis of FAB mass and ^{13}C NMR spectra consistent to the molecular formula of alkylated phenol $C_{12}H_{17}O_3$. The ^{1}H NMR spectrum of **2** showed two one-proton doublets at *δ* 7.35 (*J*=7.8 Hz) and 5.46 (*J*=7.8 Hz) assigned to *ortho*-coupled H-5 and H-6, respectively, a two-proton broad signal at *δ* 3.39 ascribed to dioxymethylene protons, other methylene protons at *δ* 2.51 (2H) and 1.22 (6H) and a three-proton triplet at *δ* 0.83 (*J*=6.1 Hz) accounted to C-11 primary methyl protons. The ^{13}C NMR spectrum of **2** exhibited signals for aromatic carbons between *δ* 164.06-140.13, dioxymethylene carbon at *δ* 100.05, other methylene carbons at 31.73 (C-7), 28.68 (C-8, C-9) and 23.96 (C-10) and methyl carbon at *δ* 14.26 (C-11). On the basis of these evidences the structure of **2** been estabilished as *n*-pentanyl-2,3-dioxymethylene-4-hydroxybenzene. This is a new phenolic derivative.



2

Compound **3**, designated as *n*-decanyl-2,3-(methylenedioxy)phenol, was obtained as a colourless crystalline mass from chloroform-methynol (19:1) eluents. It gave positive tests for phenols, showed UV absorption maximum at 275 nm and displayed IR absorption bands for hydroxyl group (3398 cm^{-1}) and aromatic ring (1541, 1526, 930 cm^{-1}). It had a molecular ion peak at *m/z* 279 $[M+H]^+$ ($C_{17}H_{27}O_3$) established on the basis of FAB mass and ^{13}C NMR spectra. An ion peak arising at *m/z* 137 [C_1-C_7 fission, $C_6H_2(OH)(OCH_2O)]^+$ indicated that methylenedioxyphenol was linked to decanyl chain. The ^1H NMR spectrum of **3** showed two one-proton doublets at δ 7.37 (*J*=7.9 Hz) and 5.47 (*J*=7.9 Hz) assigned to aromatic *ortho*-coupled H-5 and H-6, respectively. A two-proton signal at δ 3.37 was ascribed to dioxymethylene protons. The other methylene protons appeared as a two-proton triplet at δ 2.41 (*J*=7.1, H_2-7), as a multiplet at δ 1.84 (H_2-8) and as a broad signal at δ 1.22 (14H). A three-proton triplet at δ 0.85 (*J*=6.1 Hz) was accounted to primary C-16 methyl protons. The ^{13}C NMR spectrum of **3** displayed signals for aromatic carbons between δ 164.59-137.91, dioxymethylene carbon at δ 100.39, other methylene carbon between δ 33.81-22.45 and methyl carbon at δ 14.26 (C-16). On the basis of above discussion, the structure of **3** has been formulated as *n*-decanyl-2,3-dioxymethylene-4-hydroxybenzene. This is also anew phenolic derivative.

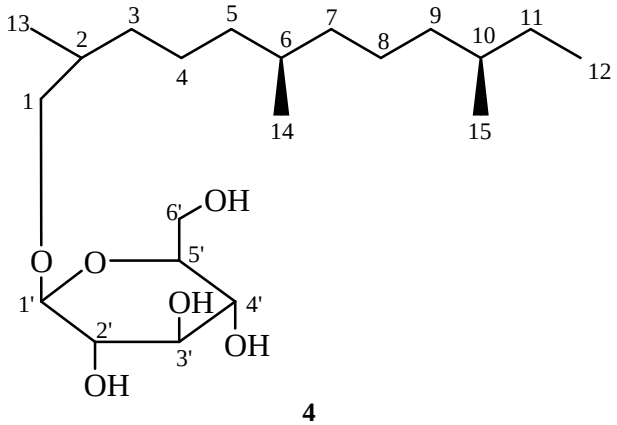


3

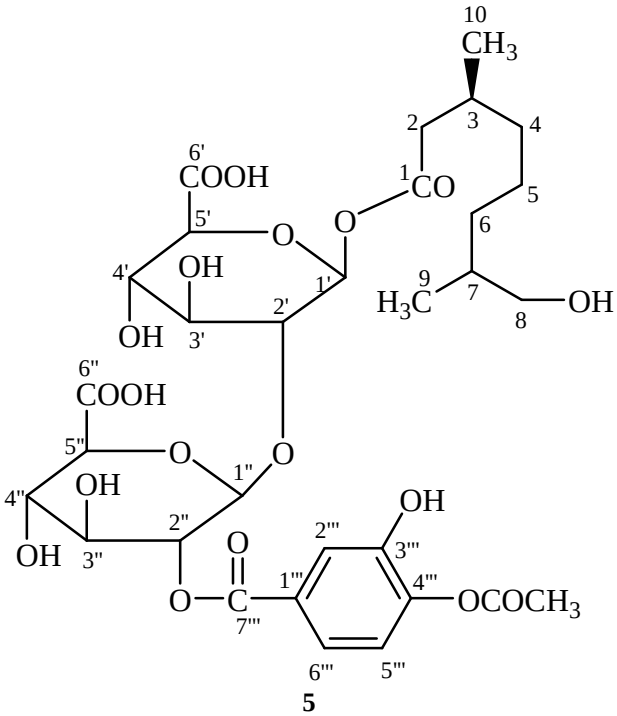
Compound **4**, named dihydrocitronellyl glucoside, was obtained as colourless crystals from chloroform-methanol (9:1) eluents. It gave positive tests of glucosides and showed IR absorption band for hydroxyl groups (3450, 3285 cm^{-1}). It had a molecular ion peak at *m/z* 381 $[M+H]^+$ ($C_{21}H_{33}O_6$) on the basis of FAB mass and ^{13}C NMR spectra. The ion peaks arising at *m/z* 217 $[M-C_6H_{11}O_5]^+$ and 200 $[M-C_6H_{12}O_6]^+$ suggested the location of a C_6 sugar unit to the monoterpene. The ^1H NMR spectrum of **4** showed a one-proton doublet at δ 5.12 (*J*=7.2 Hz) assigned anomaric H-1′ protons, other sugar protons as one-proton multiplets at δ 4.25 (C-5′), 3.98 (H-2′), 3.56 (H-3′) and 3.43 (H-4′) and doublets at δ 3.13 (*J*=6.1 Hz, H_2-6′a) and 3.10 (*J*=6.1 Hz, H_2-6′b) and oxygenated H_2-1 methylene protons as doublets at δ 3.39 (*J*=8.1 Hz) and 3.35 (*J*=6.3 Hz). Three doublets at δ 1.23 (*J*=6.3 Hz), 0.96 (*J*=6.0 Hz) and 0.92 (*J*=6.7 Hz) and a triplet at δ 0.83 (*J*=6.3 Hz), all integrated for three-protons each, were attributed to secondary C-13, C-14 and C-15 and primary C-12 methyl protons, respectively, all attached to saturated carbons. The other methine and methylene protons appeared between δ 2.50-1.29. The ^{13}C NMR spectrum of **4** displayed signals for anomeric carbon at δ 103.53 (C-1′), other sugar carbons between δ 78.70-63.46, oxygenated methylene carbon at δ 65.51 (C-1) and methyl carbons at δ 13.34 (C-12), 28.23 (C-13), 24.94 (C-14) and 21.62 (C-15). Acid hydrolysis of **4** yielded D-glucose as a glycone unit. On the basis of these evidences the structure of **4** has been elucidated as 2,6,10-trimethyl dodecan-1-ol-1β-D-glucopyranoside. This is a new monoterpenic glucoside.

Compound **5**, dihydrocitronelloyl glucuronosidic ester, was obtained from chloroform-methanol (9:1) eluents. It showed test for glycosides and had IR absorption bands for hydroxyl groups (3450, 3310 cm^{-1}), ester groups (1735, 1721 cm^{-1}), carboxylic functions (1690 cm^{-1}) and aromatic ring (1560, 950 cm^{-1}). Its molecular ion peak was determined at *m/z* 719 $[M+H]^+$ ($C_{31}H_{43}O_{19}$) on the basis of FAB mass and ^{13}C NMR spectra. The ion peaks arising at *m/z* 171 (C_1-O fission, $C_{10}H_{19}O_2)^+$ and 187 [$C_{1'}$-O fission, $C_{10}H_{19}O_3]^+$ indicated the location of dihydrocitronelloyl unit to the glycosidic unit. The ion fragments generating at *m/z* 179 [$C_{7'''}$-O fission, HO-C_6H_3 $(OCOCH_3)CO]^+$ and 523 $[M-179]^+$ suggested the existence of acetyl dihydroxybenzoyl group linked to the sugar unit. The ^1H NMR spectrum of **5** showed three one-proton aromatic proton signals as doublets at δ 7.79 (*J*=2.5 Hz) and 7.71 (8.9 Hz) and as a double doublet at δ 7.58 (2.5, 8.9 Hz) and assigned to meta-coupled H-2‴, ortho-coupled H-5‴ and ortho, meta-coupled H-6‴ protons, respectively, two one-proton doublets at δ 5.32 (*J*=7.1 Hz) and 5.21 (*J*=7.3 Hz) ascribed to anomeric H-1′, and H-1″ protons, respectively, other sugar proton signals between δ 4.23-3.91 and a

two-proton doublet at δ 3.13 (J=6.5 Hz) attributed to hydroxyl methylene H_2-8 protons. Three signals as a broad signal at δ 2.50 and as doublets at δ 1.23 (J=6.5 Hz) and 0.86 (J=6.1 Hz), all integrated for three protons each, were accounted correspondingly to acetyl and to secondary C-9 and C-10 methyl protons. The ^{13}C NMR spectrum of **5** exhibited signals for carboxylic carbons at δ 179.43 (C-6′) and 177.20 (C-6″), ester carbons at δ 175.42 (C-1), 173.77 (C-7‴) and 171.92 (Ac), anomeric carbons from δ 105.23 (C-1′) and 101.61 (C-1″), other sugar carbons between δ 79.36-64.07, aromatic carbons from δ 157.76-115.36 and hydroxyl methylene carbon at δ 60.78 (C-8). The presence of H-2″ and H-2′ signals in the deshielded region at 4.23 and 4.17, respectively, in the ^{1}H NMR spectrum and carbon signals at δ 79.36 (C-2″) and 71.53 (C-2′) suggested (2′→1″) linkeage of sugar units and location of the benzoyl group at C-2″. Acid hydrolysis of **5** yielded glucuronic acid as a glycone residue. On the basis of the aforementioned description the structure of **5** has been estabilished as 8-hydroxydihydrocitronelloyl-1β-D-glucuronosyl-(2′→1″)-β-D-glucuronopyranosyl-2″-(3‴-hydroxy-4‴-acetoxy) benzoate. This is a new monoterpenic diglucuronosyl ester.



4



5

CONCLUSION

Five new phytoconstituents have been isolated from the silk cocoons of *Bombyx mori* which may be responsible for the medicinal properties of drug.

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REFERENCES

[1] Anonymous; Wealth of India. A dictionary of Raw Materials and Industrial Product, National Institute of Science Communication and Information Resources, CSIR, New Delhi, **2003**, vol 9, p.327-353.
[2] Takasu Y; Yamada H; Tsubouchi K. *Biosci Biotech Biochem*, **2002**, 66(12), 2715-2718.
[3] C Hirayama; H Ono; Y Tamura; M Nakamura. *Phytochemistry*, **2006**, 67(6), 579-583.
[4] H-Y Wang; Y-J Wang; L-X Zhou; L Zhu; Y-Q Zhang. *Food Funct*, **2012**, 3, 150-158.
[5] N Fujimoto and K Kawakami. *J Seric Sci Japan*, **1958**, 27, 391-392.
[6] K Hayashiya; S Sugimoto; N Fujimoto. *J Seric Sci Japan*, **1959**, 28, 27-29.
[7] Y Tamura; K Nakajima; K Nagayasu; C Takabayashi. *Phytochemistry*, **2002**, 59 (3), 275-278.
[8] C Hirayama; H Ono; Y Tamura; K Kanno; M Nakamura. *Phytochemistry*, **2008**, 69, 1141-1149.
[9] S Sridhara and JV Bhat. *J Insect Physiol*, **1965**, 11(4), 449-462.
[10] M Lahtinen; L Kapari; J Kentta. *J Chem Ecol*, **2006**, 32(3), 537-46.
[11] JP Salminen; M Lahtinen; K Lempa; L Kapari; E Haukioja; K Pihlaja. *Z Naturforsch C*, **2004**, 59(5-6), 437-444.
[12] SA Ahmad and TL Hopkins. *Insect Biochem Mol Biol*, **1993**, 23, 581-589.
[13] C Rausell; J Llorca; MD Real. *Arch Insect Biochem Physiol*, **1997**, 34(3), 347-358.
[14] R Tanaka; T Hanashima; H Yamada. *Biosci Biotechnol Biochem*, **2010**, 74(10), 2133-2136.
[15] GX Zhou; JW Ruan, MY Huang; WC Ye; YW He. *Zhong Yao Cai*, **2007**, 30(11): 1384-1385.
[16] AK Dixit; K Kadavul; S Rajalakshmi; MS Shekhawat. *Indian J Traditional Knowledge*, **2010**, 9(1), 116-118.
[17] HM Said. Hamdard Pharmacopoeia of eastern medicine, The Times Press, Sadar, Karachi. **1970**, p. 41-57.