Novel Glycosides from Noni (*Morinda citrifolia*)

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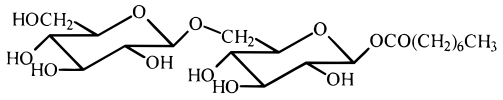
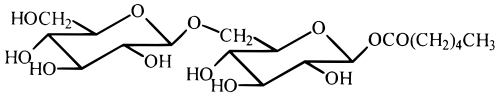
Three new glycosides were isolated from the fruits of noni (*Morinda citrifolia). Their structures were determined to be* 6-*O*-(*β*-D-glucopyranosyl)-1-*O*-octanoyl-*β*-D-glucopyranose (**1**), 6-*O*-(*β*-D-glucopyranosyl)-1-*O*-hexanoyl-*β*-D-glucopyranose (**2**), and 3-methylbut-3-enyl 6-*O*-*β*-D-glucopyranosyl-*β*-D-glucopyranoside (**3**) using MS and NMR methods.

Noni, the common name for *Morinda citrifolia* L. (Rubiaceae) is a plant typically found in the Tahitian and Hawaiian islands.[1] The bark, stem, root, leaf, and fruit have been used traditionally for many diseases, including diabetes, hypertension, and cancer,[2,3] and are all mentioned as Hawaiian herbal remedies. In the earlier pharmacological work, the juice of noni fruits was found to prolong the life of mice implanted Lewis lung carcinoma.[2] Extracts of noni roots have been tested for their analgesic effects and have shown a significant, dose-related, central analgesic activity in treated mice.[4] The chemical components of noni have not been well studied, and only several anthraquinones and asperuloside were previously isolated.[1,5−9] Here we report the isolation of three novel glycosides (**1**−**3**) from the *n*-BuOH-soluble fraction of noni fruit extract.



1



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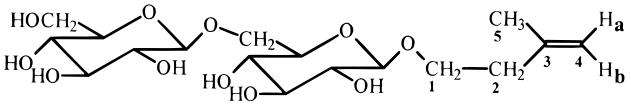

3

Compound **1** was obtained as a white powder. The negative APCIMS exhibited a pseudomolecular ion peak at *m*/*z* 467 [M − 1]$^-$ and the positive APCIMS showed a significant pseudomolecular ion peak at *m*/*z* 486 [M + NH_4]$^+$, compatible with the molecular formula $C_{20}H_{36}O_{12}$. In the ^{1}H and ^{13}C NMR, compound **1** showed signals consistent with an octanoyl partial structure. In the ^{1}H NMR spectrum of **1**, two anomeric proton signals at *δ* 5.45 (1H, d, *J* = 7.8 Hz) and 4.31 (1H, d, *J* = 7.8 Hz) were observed. The ^{13}C NMR also displayed signals at *δ* 104.5 (d), 77.9 (d), 77.9 (d), 75.0 (d), 71.4 (d), and 62.6 (t), attributable to terminal *β*-D-glucose,[10] and signals at *δ* 95.5 (d), 77.7 (d), 77.7 (d), 73.8 (d), 70.8 (d), and 69.4 (t) for the inner glucose. Comparison with literature values indicated 1→6 linkage of these two glucose units,[11] and the octanoyl moiety was placed on the anomeric carbon of the central glucose. The above evidence established the structure of **1** as 6-*O*-(*β*-D-glucopyranosyl)-1-*O*-octanoyl-*β*-D-glucopyranose.

The structure of **1** was confirmed by ^{1}H−^{1}H COSY, NOESY, HMQC, and HMBC spectra. HMBC experiments showed correlation contours between H-1 of the central glucose (*δ* 5.45) and the carbonyl carbon of the octanoyl moiety (*δ* 174.1), and between H-1 of the terminal glucose (*δ* 4.31) and C-6 of the central glucose (*δ* 69.4).

Compound **2** was also obtained as a white powder. The negative APCIMS exhibited a significant pseudomolecular ion peak at *m*/*z* 439 [M − 1]$^-$, and the positive APCIMS showed an ion peak at *m*/*z* 458 [M + NH_4]$^+$. These MS data together with the ^{1}H and ^{13}C NMR data, suggested the molecular formula $C_{18}H_{32}O_{12}$. The IR spectrum showed hydroxyl and carbonyl absorptions. In the ^{1}H NMR spectrum, **2** showed signals similar to those of **1**. Only slight differences were observed in the high field where, instead of the signals for an octanoyl moiety, signals for a hexanoyl moiety were observed. This observation was further supported by the ^{13}C NMR spectrum, which showed signals at *δ* 14.3 (q), 23.4 (t), 25.3 (t), 32.3 (t), 34.8 (t), and 174.1 (s), assignable to a hexanoyl moiety.[12] The remaining ^{13}C NMR signals for the two glucose moieties were identical with those of **1**. The ^{1}H NMR signals for the two anomeric protons were observed at *δ* 5.45 and 4.31. Analysis of the ^{1}H−^{1}H COSY, HMQC, and HMBC spectra led to assignment of all ^{1}H and ^{13}C NMR signals for **2**. Thus, **2** was identified as 6-*O*-(*β*-D-glucopyranosyl)-1-*O*-hexanoyl-*β*-D-glucopyranose.

Compound **3** exhibited a significant pseudomolecular ion peak at *m*/*z* 409 [M − 1]$^-$ in negative APCIMS and an ion peak at 428 [M + NH_4]$^+$ in the positive APCIMS. MS data together with the ^{1}H and ^{13}C NMR data suggested molecular formula $C_{17}H_{30}O_{11}$. The ^{1}H and ^{13}C NMR spectra of **3** clearly showed the signals for a 1→6-linked *β*-D-glucopy-

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ranosyl-β-D-glucopyranose moiety.[11] In addition to signals for sugars, the ^{1}H NMR spectrum showed the presence of one methyl (δ 1.75), two methylenes [δ 2.35 (2H), and 3.65 (1H), 3.99 (1H)], and one exomethylene [δ 4.74 (1H) and 4.75 (1H)], while the remaining ^{13}C signals were observed at δ 23.0 (q), 38.7 (t), 69.5 (t), 112.1 (t), and 143.9 (s). These data were assignable to the partial structure CH_2═C(CH_3)-CH_2CH_2O−.

Analysis of the ^{1}H−^{1}H COSY, HMQC, and HMBC spectra led to the assignment of the ^{1}H and ^{13}C NMR data of **3** and confirmed the CH_2═C(CH_3)CH_2CH_2O− moiety. A ^{13}C signal at 112.1 (t) in the HMQC spectrum correlated with the exomethylene proton signals (δ 4.74 and 4.75). The former signal also showed correlation with carbon signals at δ 143.9 (C-3) and 38.7 (C-2), while the latter signal showed correction with carbon signals at δ 143.9 and 23.0 (C-5) in the HMBC spectrum. The linkage of sugars and aglycon was consistent with the HMBC experiments, in which correlations were observed between H-1 of the central glucose (δ 4.27) and a CH_2 at δ 69.5. Thus, the structure of **3** was elucidated as 3-methylbut-3-enyl 6-*O*-β-D-glucopyranosyl-β-D-glucopyranoside.

Experimental Section

General Experimental Procedures. Melting points were determined on a Thomas-Hoover "Unimelt" apparatus. Optical rotations were obtained on a JASCO DIP-181 polarimeter. IR spectra were recorded on a Nicolet 5000 infrared spectrometer. ^{1}H and ^{13}C NMR spectra were obtained on a U-500 instrument (Varian Inc., Melbourne, Australia) operating at 500 and 125 MHz, respectively, and compounds were analyzed in CD_3OD with tetramethylsilane (TMS) as an internal standard. ^{1}H−^{1}H COSY, NOESY, HMQC, and HMBC were performed on a U-500 instrument (Varian Inc., Melbourne, Australia). APCI-MS was obtained on a Fisons/VG Platform II mass spectrometer. TLC was performed on Sigma-Aldrich TLC plates (250 μm thickness, 2−25 μm particle size), with compounds visualized by spraying with 5% (v/v) H_2SO_4 in ethanol solution.

Plant Material. Noni fruit was collected from Hawaii in 1999. A voucher specimen (WM199900188) was deposited in the Department of Food Science, Cook College, Rutgers University.

Extraction and Isolation. Dried noni fruits (200 g) were extracted with 95% EtOH (1 L) at room temperature for one week. The extract was concentrated to dryness under reduced pressure, the residue was suspended in H_2O (500 mL) and partitioned successively with hexanes (3 × 500 mL), ethyl acetate (3 × 500 mL), and *n*-BuOH (3 × 500 mL).

The *n*-BuOH-soluble fraction (40 g) was subjected to column chromatography on 500 g Si gel, eluted with ethyl acetate−CH_3OH−H_2O with increasing CH_3OH and H_2O (10:1:1, 6:1:1, 5:1:1, 4:1:1, 3:1:1, 2:1:0.5, each 1000 mL), and 500-mL fractions were collected. Fractions 7 and 8 were combined and subjected to a RP_{18} column, eluted by CH_3OH−H_2O (1:1) to afford pure compounds **3** (300 mg) and **2** (100 mg). Fraction 6 was first subjected to a normal-phase Si gel column, eluted with $CHCl_3$−CH_3OH−H_2O (3:1:0.1) to yield two subfractions; then subfraction 1 was subjected to a reversed-phase column, eluted by CH_3OH−H_2O (1:1), to provide **1** (200 mg).

6-*O*-(β-D-Glucopyranosyl)-1-*O*-octanoyl-β-D-glucopyranose (1): white powder (200 mg); mp 150−152 °C; $[\alpha]_D$ +24.0° (*c* 0.13 MeOH); IR (film) ν_{max} 3404 (OH), 2928, 2857, 1740 (CO), 1170, 1072 cm^{-1}; ^{1}H NMR (CD_3OD, 500 MHz) δ 5.45 (1H, d, *J* = 7.8 Hz, H-1′), 4.31 (1H, d, *J* = 7.8 Hz, H-1″), 4.14 (1H, dd, *J* = 11.4, 3.0 Hz, H-6′), 3.85 (1H, dd, *J* = 12.1, 2.2 Hz, H-6″), 3.76 (1H, dd, *J* = 11.4, 5.1 Hz, H-6′), 3.65 (1H, dd, *J* = 12.1, 5.4 Hz, H-6″), 3.53 (1H, m, H-5′), 3.42 (1H, m, H-4′), 3.41 (1H, m, H-3′), 3.34 (1H, dd, *J* = 9.1, 8.8 Hz, H-3″), 3.32 (1H, dd, *J* = 9.3, 7.8 Hz, H-2′), 3.28 (1H, dd, *J* = 8.8, 8.8 Hz, H-4″), 3.25 (1H, m, H-5″), 3.20 (1H, dd, *J* = 9.1, 7.8 Hz, H-2″), 2.38 (2H, m, H-2), 1.62 (2H, tt, *J* = 7.3 Hz, H-3), 1.30−1.35 (8H, m, H-4, H-5, H-6, H-7), 0.90 (3H, t, *J* = 7.1 Hz, H-8); ^{13}C NMR (CD_3OD, 125 MHz) δ 174.1 (s, C-1), 104.5 (d, C-1″), 95.5 (d, C-1′), 77.9 (d, C-3″), 77.9 (d, C-5″), 77.7 (d, C-3′), 77.7 (d, C-5′), 75.0 (d, C-2″), 73.8 (d, C-2′), 71.4 (d, C-4″), 70.8 (d, C-4′), 69.4 (t, C-6′), 62.6 (t, C-6″), 34.9 (t, C-2), 32.8 (t, C-6), 30.1 (t, C-4), 30.1 (t, C-5), 25.6 (t, C-3), 23.7 (t, C-7), 14.4 (q, C-8); negative APCIMS *m*/*z* 467 [M − 1]$^-$ and positive APCIMS *m*/*z* 486 [M + NH_4]$^+$; HRFABMS [M + 1]$^+$ *m*/*z* 469.2279 (calcd for $C_{20}H_{37}O_{12}$, 469.228).

6-*O*-(β-D-Glucopyranosyl)-1-*O*-hexanoyl-β-D-glucopyranose (2): white powder (100 mg); mp 147−148 °C; $[\alpha]_D$ +8.3° (*c* 0.24, MeOH); IR (film) ν_{max} at 3394 (OH), 2943, 2861, 1749 (CO), 1471, 1379 and 1072 cm^{-1}; ^{1}H NMR (CD_3OD, 500 MHz) δ 5.45 (1H, d, *J* = 7.8 Hz, H-1′), 4.31 (1H, d, *J* = 7.8 Hz, H-1″), 4.14 (1H, dd, *J* = 11.5 2.2 Hz, H-6′), 3.85 (1H, dd, *J* = 12.0, 2.2 Hz, H-6″), 3.76 (1H, dd, *J* = 11.5, 5.1 Hz, H-6′), 3.65 (1H, dd, *J* = 12.0, 5.4 Hz, H-6″), 3.53 (1H, m, H-5′), 3.42 (1H, m, H-4′), 3.41 (1H, m, H-3′), 3.34 (1H, dd, *J* = 9.1, 8.3 Hz, H-3″), 3.32 (1H, m, H-2′), 3.28 (1H, dd, *J* = 8.3, 8.3 Hz, H-4″), 3.26 (1H, m, H-5″), 3.20 (1H, dd, *J* = 9.3, 7.8 Hz, H-2″), 2.39 (2H, m, H-2), 1.62 (2H, tt, *J* = 7.3 Hz, H-3), 1.34 (4H, m, H-4, H-5), 1.34 (2H, m, H-5), 0.91 (3H, t, *J* = 7.1 Hz, H-6); ^{13}C NMR (CD_3OD, 125 MHz) δ 174.1 (s, C-1), 104.5 (d, C-1″), 95.5 (d, C-1′), 78.0 (d, C-5″), 77.9 (d, C-3″), 77.8 (d, C-3′), 77.7 (d, C-5′), 75.0 (d, C-2″), 73.9 (d, C-2′), 71.5 (d, C-4″), 70.8 (d, C-4′), 69.4 (t, C-6′), 62.7 (t, C-6″), 34.8 (t, C-2), 32.3 (t, C-4), 25.3 (t, C-3), 23.4 (t, C-5), 14.3 (q, C-6); negative APCIMS *m*/*z* 439.0 [M − 1]$^-$ and positive APCIMS *m*/*z* 458 [M + NH_4]$^+$; HRFABMS [M + 1]$^+$ *m*/*z* 441.1966 (calcd for $C_{18}H_{33}O_{12}$, 441.1972).

3-Methylbut-3-enyl 6-*O*-β-D-glucopyranosyl-β-D-glucopyranoside (3): white powder (300 mg); mp 78−80 °C; $[\alpha]_D$ +39.0° (*c* 0.11, MeOH); IR (film) ν_{max} 3404 (OH), 2923, 1652, 1384, 1166, 1046 cm^{-1}; ^{1}H NMR (CD_3OD, 500 MHz) δ 4.75 (1H, m, H-4b), 4.74 (1H, m, H-4a), 4.37 (1H, d, *J* = 7.8 Hz, H-1″), 4.27 (1H, d, *J* = 7.8 Hz, H-1′), 4.14 (1H, dd, *J* = 11.5, 1.9 Hz, H-6′), 3.99 (1H, dt, *J* = 9.8, 7.1 Hz, H-1), 3.86 (1H, dd, *J* = 11.9, 1.8 Hz, H-6″), 3.78 (1H, dd, *J* = 11.5, 5.7 Hz, H-6′), 3.66 (1H, dd, *J* = 11.9, 5.4 Hz, H-6″), 3.65 (1H, dt, *J* = 9.8, 7.1 Hz, H-1), 3.45 (1H, ddd, *J* = 9.5, 5.7, 1.9 Hz, H-5′), 3.35 (1H, m, H-3′), 3.34 (1H, m, H-3″), 3.34 (1H, m, H-4′), 3.28 (1H, dd, *J* = 8.3, 8.3 Hz, H-4″), 3.26 (1H, m, H-5″), 3.21 (1H, dd, *J* = 9.0, 7.8 Hz, H-2″), 3.17 (1H, dd, *J* = 9.3, 7.6 Hz, H-2′), 2.35 (2H, t, *J* = 7.1 Hz, H-2), 1.75 (3H, br s, H-5); ^{13}C NMR (CD_3OD, 125 MHz) δ 143.9 (s, C-3), 112.1 (t, C-4), 104.8 (d, C-1″), 104.4 (d, C-1′), 78.0 (d, C-3″), 78.0 (d, C-5″), 77.9 (d, C-3′), 77.0 (d, C-5′), 75.1 (d, C-2″), 75.0 (d, C-2′), 71.5 (d, C-4″), 71.4 (d, C-4′), 69.8 (t, C-6′), 69.5 (t, C-1), 62.7 (t, C-6″), 38.7 (t, C-2), 23.0 (q, C-5); negative APCIMS *m*/*z* 409 [M − 1]$^-$ and positive APCIMS 428 [M + NH_4]$^+$; HRFABMS [M + 1]$^+$ *m*/*z* 411.1869 (calcd for $C_{17}H_{31}O_{11}$, 411.1866).

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