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Chemical Constituents of Fermented Noni (*Morinda citrifolia*) Juice Exudates and Their Biological Activity

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Abstract – In a continuing study of the fermented noni (*Morinda citrifolia*) juice exudates, five compounds, heptanyl 2-*O*-*β*-D-xylofuranosyl-(1 → 6)-*β*-D-glucopyranoside (**1**), *n*-butyl *β*-D-glucopyranoside (**2**), (1*S*)-(3-ethenyl-phenyl)-1,2-ethanediol (**3**), (2*S*)-2-hydroxybutanedioic acid (**4**), and daucosterol (**5**) were isolated from the buthanol partition of the extract. The structures of the isolates were identified by 1D and 2D NMR, and MS experiments, as well as by comparison of their data with the published values. Among the isolates, compounds **1** – **3** were isolated for the first time from the plant species. The isolated compounds were evaluated for their cancer chemopreventive potential based on their ability to inhibit nitric oxide (NO) production and tumor necrosis factor alpha (TNF-α)-induced NF-κB activity, and quinonone reductase-1 (QR1)-inducing effect.
Keywords – *Morinda citrifolia*, Rubiaceae, alkyl disaccharide, phenyl ethandiol, anti-inflammatory activity

Introduction

Noni (*Morinda citrifolia* L., Rubiaceae) is a small evergreen shrub or tree growing in tropical and subtropical areas worldwide. Originally native to Southeastern Asia, the noni plant was spread to Australia, Hawaii, French Polynesia Islands, and other tropical areas. Noni fruits were traditionally used for the improvement of various health problems, such as cancer, infection, arthritis, diabetes, asthma, and pain.[1,2] Recent study has shown that noni fruits possess antioxidant, anti-inflammatory, liver-protective, and immunomodulatory effects.[3,4] So far, many of secondary metabolites, such as flavonoids, lignans, iridoids, coumarins, anthraquinones, polysaccharides, terpenoids, sterol, and fatty acid glycosides have been isolated from this species.[5-11]

In a previous research, we have reported the isolation and structural elucidation of iridoid glycosides and fatty acid ester disaccharide, along with phenolic compounds from the fermented juice of noni fruits with their biological activity.[12]

As part of a continuing search for anti-inflammatory agents, two alkylated saccharides, a phenolic compound, adioic acid, and a steroid glycoside have been isolated from the butanol partition of fermented juice of *M. citrifolia* fruits. This paper reports the isolation and structure elucidation of compounds **1** - **5** (Fig. 1), as well as their biological activity.

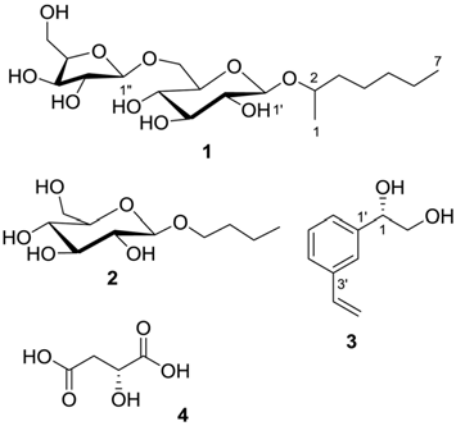

Fig. 1. Structures of compounds **1** - **4** isolated from the fermented noni juice.

Experimental

General – Optical rotations were measured on a Rudolph Research Autopol IV multi wave length polarimeter. UV spectra were scanned on a Shimadzu

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PharmaSpec-1700 UV-visible spectrophotometer. IR spectra were obtained on a Bruker Tensor-27 spectrometer. 1D and 2DNMR spectra were recorded on a Bruker AVANCE (400 MHz) NMR spectrometer. LCMS spectra were obtained with an Agilent 6530 LC-qTOF high mass accuracy mass spectrometer operated in both the positive- and negative-ion modes. Thin-layer chromatography (TLC) was performed on silica gel 60 F_{254} (0.25 mm, Merck, Darmstadt, Germany). Silica gel (230 - 400 mesh, Merck) and RP-18 (YMC·GEL ODS-A, 12 nm, S-150 μm, YMC, Tokyo, Japan) were used for column chromatography. Semi-preparative HPLC was conducted on a Beckman Coulter Gold-168 system equipped with a DAD detector, utilizing an Alltech reversed-phase Econosil C_{18} column (10 μm, 10 × 250 mm) with a flow rate of 2 mL/min.

Plant material – The fruits of *M. citrifolia* were harvested from a commercial orchard in Kalapana, Hawai'i, USA from 2012 through 2014. The plant was identified by Dr. Marisa M. Wall, United States Department of Agriculture, Daniel K. Inouye U.S. Pacific Basin Agricultural Research Center, Hilo, USA. Fruits harvested were at the firm, yellow-white maturity stage. The fruits were washed with soft-bristled brushes in soapy water, rinsed in tap water, air-dried, sorted, and placed into sterile 3.8 L wide-mouth glass jars (~1.5 to 2.0 kg fruit per jar) with lids, and stored in the dark at 22 ºC in a walk-in chamber for 16 days. During this time, juice exuded from the fruit and the naturally fermented juice were drained from containers by pouring through a sterile plastic sieve into a metal pot, and pasteurized at 85 ºC for 3 min; the pasteurized juice was poured immediately into 3 L food-grade sterile buckets with lids and frozen at −10 ºC. A voucher specimen (no. FNJ 005) was deposited at the Natural Product Chemistry Laboratory, Daniel K. Inouye College of Pharmacy, University of Hawai'i at Hilo.

Extraction and isolation – The freeze-dried fermented juice from nonifruits (10 L) was extracted with ethyl acetate (EtOAc) and *n*-butanol (3 × 4 L), successively. The butanol-soluble partition (80.0 g) was subjected to silica gel column chromatography (CC; ϕ20 cm; 230 - 400 mesh, 5 kg) eluted by $CHCl_3$-MeOH (100:1 to 50:50) to afford 12 fractions (BS1BS12).Fraction BS2 (0.5 g) was chromatographed on a RP-18 gel (ϕ2 × 100 cm; 40 - 63 mesh, 200 g) column, with H_2O-MeOH (100:0 to 70:30) as the solvent system, yielding five subfractions (BS2R1 to BS2R5). Fraction BS2R3 was subjected to semi-preparative HPLC [MeOH/H_2O (0:100 to 80:20)] to yield compound **3** (0.5 mg, t_R97 min). Compound **2** (100 mg) was obtained by recrystallization using methanol from fraction BS10. Fraction BS11 (0.8 g) was subjected to passage over a column containing RP-18 gel (ϕ3 × 100 cm; 40 - 63 mesh, 400 g), with H_2O-MeOH (100:0 to 30: 70) as the solvent system, yielding 10subfractions (BS11R1 to BS11R10). Compounds **1** (15 mg) and **4** (5 mg) were purified by repeated sephadex LH 20 (300 g) CC and semi-preparative HPLC [MeOH/H_2O (0:100 to 80:20)] from fractions BS11R2 and BS11R5, respectively. Daucosterol, **5** (8 mg) was obtained by recrystallization from fraction BS12.

Heptanyl 2-*O*-β-D-xylofuranosyl-(1→6)-β-D-glucopyranoside (1) – White amorphous powder, IR ν_{max} (KBr) 3320, 1732 cm^{-1}; LCMS *m/z*411 $[M + H]^+$; ^{1}H-NMR (400 MHz, MeOD) δ_H 3.83, (1H, m, H-2), 1.63 (1H, m, H-3a), 1.40 (1H, m, H-3b), 1.37 (2H, m, H-4), 1.37 (2H, m, H-6), 1.33 (2H, m, H-5),1.24 (3H, d, *J* = 6.4 Hz,CH_3-1),0.93 (3H, t, *J* = 7.0 Hz, CH_3-7), 4.33 (1H, d, *J* = 8.0 Hz, H-1'), 3.21 (1H, m, H-2'), 3.30 (1H, m, H-3'), 3.34 (1H, m, H-4'), 3.42 (1H, m, H-5'), 4.07 (1H, dd, *J* = 11.2, 2.0 Hz, H-6'a), 3.76 (1H, dd, *J* = 11.2, 5.6 Hz, H-6'b), 4.35 (1H, d, *J* = 7.6 Hz, H-1"), 3.17 (1H, m, H-2"), 3.34 (1H, m, H-3"), 3.50 (1H, m, H-4"), 3.88 (1H, dd, *J* = 11.2, 6.0 Hz, H-5"a), 3.19 (1H, overlapped, H-5"b); ^{13}C-NMR (100 MHz, MeOD) δ_C 76.3 (C-2), 36.3 (C-3), 31.7 (C-5), 24.6 (C-4), 22.2 (C-6), 20.6 (CH_3-1), 13.0 (CH_3-7), 102.6 (C-1'), 73.4 (C-2'), 76.2 (C-3'), 70.0 (C-4'), 75.4 (C-5'), 68.3 (C-6'), 104.0 (C-1"), 73.8 (C-2"), 76.5 (C-3"), 69.7 (C-4"), 65.5 (C-5").

***n*-Butyl β-D-glucopyranoside (2)** – Amorphous colorless crystal, IR ν_{max} (KBr) 3345, 2842 cm^{-1}; LCMS *m/z*237 $[M + H]^+$; ^{1}H-NMR (400 MHz, MeOD) δ_H 3.53 (2H, m, H-1), 1.58 (2H, m, H-2), 1.42 (2H, m, H-3), 0.96 (3H, t, *J* = 7.4 Hz, CH3-4), 3.74 (1H, d, *J* = 8.0 Hz, H-1'), 3.65-3.76 (4H, m, H-2' to H-5'), 3.93 (1H, d, *J* = 9.6 Hz, H-6'a), 3.70 (1H, dd, *J* = 12.4, 2.0 Hz, H-6'b); 13C-NMR (100 MHz, MeOD) δ_C 60.2 (C-1), 31.9 (C-2), 19.0 (C-3), 12.9 (C-4), 100.2 (C-1'), 69.2 (C-2'), 69.6 (C-3'), 63.7 (C-4'), 70.1 (C-5'), 62.0 (C-6').

(1*S*)-(3-Ethenyl-phenyl)-1,2-ethanediol (3) – Colorless oil, $[\alpha]_D^{25}$ +32 (*c* 0.12, MeOH), UV (MeOH) λ_{max} (log ε) 268 (4.1) nm; LCMS *m/z*165 $[M + H]^+$; ^{1}H-NMR (400 MHz, MeOD) δ_H 7.28 - 7.37 (3H, m, overlapped, H-4' to H-6'), 7.42 (1H, br s, H-1'), 6.77 (1H, dd, *J* = 18.0, 10.8 Hz, H-7'), 5.81 (1H, dd, *J* = 17.6, 0.8 Hz, H-8'b), 5.25 (1H, dd, *J* = 10.8, 0.8 Hz, H-8'a), 4.70 (1H, dd, *J* = 7.2, 4.8 Hz, H-1), 3.65 (1H, m, H-2); ^{13}C-NMR (100 MHz, MeOD) δ_C 143.0 (C-1), 140.0 (C-3), 135.1 (C-7'), 124.5 (C-5'), 124.2 (C-6'), 124.0 (C-4'), 111.9 (C-8'), 73.0 (C-1), 66.1 (C-2).

(2*S*)-2-Hydroxybutanedioic acid (4) – White amorphous powder, $[\alpha]_D^{25}$ −28 (*c* 0. 25, MeOH), ^{1}H-NMR (400 MHz, MeOH) δ_H 4.50 (1H, dd, *J* = 7.6, 4.4 Hz, H-2), 2.82 (1H,

dd, J = 16.0, 4.4 Hz, H-3b), 2.66 (1H, dd, J = 16.0, 8.0 Hz, H-3a); ^{13}C-NMR (100 MHz, MeOD) δ_C 175.1 (C-1), 172.8 (C-4), 67.0 (C-2), 38.6 (C-3).

Daucosterol (5) – White amorphous powder, ^{1}H-NMR (400 MHz, pyridine-d_5) δ_H 5.35 (1H, d, J = 4.8 Hz, H-6), 5.05 (1H, d, J = 7.8 Hz, H-1'), 3.91-4.59 (5H, m, sugar), 1.01 (3H, s, CH_3-19), 0.95 (3H, d, J = 6.5 Hz, CH_3-21), 0.90 (3H, d, J = 6.5 Hz, CH_3-26), 0.88 (3H, t, J = 6.6 Hz, CH_3-29), 0.86 (3H, m, J = 6.6 Hz, CH_3-28), 0.67 (3H, s, CH_3-18); ^{13}C-NMR (100 MHz, pyridine-d_5) δ_C 37.2 (C-1), 30.6 (C-2), 78.5 (C-3), 39.7 (C-4), 141.3 (C-5), 122.3 (C-6), 32.5 (C-7), 32.4 (C-8), 50.7 (C-9), 37.3 (C-10), 21.6 (C-11), 39.7 (C-12), 42.9 (C-13), 57.2 (C-14), 24.9 (C-15), 28.9 (C-16), 56.6 (C-17), 12.3 (C-18), 19.8 (C-19), 36.8 (C-20), 19.6 (C-21), 34.6 (C-22), 26.8 (C-23), 46.4 (C-24), 29.9 (C-25), 20.3 (C-26), 19.4 (C-27), 23.8 (C-28), 12.5 (C-29), 103.0 (C-1'), 75.7 (C-2'), 78.8 (C-3'), 72.1 (C-4'), 79.0 (C-5'), 63.2 (C-6').

Tumor necrosis factor-α (TNF-α) activated nuclear factor-kappa B (NF-κB) assay – Human embryonic kidney cells 293 Panomic (Fremont, CA) were employed for monitoring changes occurring along the NF-κB pathway.[13] Stable constructed cells were seeded into 96-well plates at 20×10^3 cells per well. Cells were maintained in Dulbecco's modified Eagle's medium (DMEM) (Invitrogen Co.; Carlsbad, CA), supplemented with 10% fetal bovine serum (FBS), 100 units/mL penicillin, 100 μg/mL streptomycin, and 2 mM L-glutamine. After 48 h incubation, the medium was replaced and the cells were treated with various concentrations of test substances. TNF-α (Human, Recombinant, *E. coli*, Calbiochem, Gibbstown, NJ) was used as an activator at a concentration of 2 ng/mL (0.14 nM). The plate was incubated for 6 h. Spent medium was discarded and the cells were washed once with PBS. Cells were lysed using 50 μL (for 96-well plate) Reporter Lysis Buffer from Promega, by incubating for 5 min on a shaker, and stored at −80 °C. The luciferase assay was performed using the Luc assay system from Promega (Madison, WI). The gene product, luciferase enzyme, reacts with luciferase substrate, emitting light which was detected using a luminometer (LUMIstar Galaxy BMG). Data for NF-κB constructs are expressed as IC_{50} values (i.e. concentration required to inhibit TNF-activated NF-κB activity by 50%). As a positive control, two known NF-κB inhibitors were used: TPCK, IC_{50} = 3.8 μM and BAY-11, IC_{50} = 2.0 μM.

Inhibition of nitric oxide (NO) production in lipopolysaccharide (LPS)-activated murine macrophage RAW 264.7 cells (iNOp) assay – The level of nitrite, the stable end product of NO, was estimated as described previously.[14] Briefly, RAW 264.7 cells were seeded and incubated in 96-well culture plates at 37 °C, 5% CO_2 in a humidified air for 24 h. The cultured medium was replaced with phenol red-free medium containing various concentrations of compounds for 15 min prior to 1 μg/ml of LPS exposure for 20 h. The amount of nitrite in the culture media was measured by using Griess reagent. Under the same experimental conditions, SRB assays were performed to evaluate the cytotoxic effect of compounds toward RAW 264.7 cells. L-N^G-monomethyl arginine citrate (L-NMMA), as a positive control of this assay showed an IC_{50} value of 25.1 μM.

Quinone Reductase-1 (QR-1) Induction Assay[15] – Murine hepatoma (Hepa- 1c1c7) (CRL-2026^{TM} ATCC, Manassas, VA, USA) cells were used in this assay. Cells were plated at 200 μL at well with 0.5×10^4 cells/mL in α-MEM (minimum essential medium) without ribonucleosides or deoxyribonucleosides, supplemented with 100 units penicillin and 100 μg/mL streptomycin, and 10% FBS (Gibco). Cells were incubated for 24 h in a CO_2 incubator. After 24 h, the medium was replaced with 190 μL of fresh medium and 10 μL of test samples were added for a final concentration of 50 μM. After incubation for 48 h, digitonin was used to permeabilize cell membranes, and enzyme activity was measured by the reduction of 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide to a blue formazan. Production was measured by absorption at 595 nm. A total protein assay using crystal violet staining was run in parallel. Specific activity is defined as nmol of formazan formed/mg protein. The induction ratio of QR activity represents the specific enzyme activity of agent treated cells compared with dimethyl sulfoxide (DMSO)-treated control. The concentration to double activity (CD) was determined through a dose-response assay. 4'-Bromoflavone (CD = 0.01 μM) was used as a positive control.

Result and Discussion

Repeated chromatography of the buthanol-soluble extract from the fermented noni juice of *M. citrifolia* on silica gel and YMC-pack RP-C_{18} columns led to the isolation of five compounds (**1** - **5**) (Fig. 1).

Compound **1** was obtained as white amorphous powder. The molecular formula was evaluated as *m/z* 411 $[M + H]^+$ by the positive LCMS mass spectrometry. The IR spectrum showed one or more hydroxy group(s) and an ester carbonyl at 3320 and 1732 cm^{-1}, respectively. The ^{1}H and ^{13}C NMR and HSQC spectra of **1** showeda doublet methyl group at δ_H 1.24 (d, J = 6.4 Hz)/δ_C 20.6 (C-1), a

Table 1. Inhibition effects of compounds isolated from fermented noni juice on the TNF-α-induced NF-κB and NO production, and Quinone Reductacse-1 (QR-1) inducing activities

Compounds	Nitrite assay			NF-κB			QR1
	% inhib.[a]	% surv.[b]	IC_{50} (μM)	% inhib.[c]	% surv.[d]	IC_{50} (μM)	IR
1	5.3 ± 1.3	103.4 ± 2.8	nd[e]	56.0 ± 2.1	87 ± 4.1	nd	0.8
2	13.3 ± 1.3	79.6 ± 2.8	nd	0	107.6 ± 2.8	nd	1.4
3	3.7 ± 0.1	104.2 ± 0.4	nd	28.0 ± 2.0	88.0 ± 4.0	nd	1.8
4	–[f]	–	–	–	–	–	–
5	–	–	–	–	–	–	–
L-NMMA[g]			25.1 ± 2.3				
TPCK[h]						3.8 ± 0.6	
BAY-11[h]						2.0 ± 0.3	

[a] % inhibition of NO at a concentration of 50 μM, [b] % survival at a concentration of 50 μM, [c] % inhibition of NF-κB at 50 μM, [d] % survival at concentration of 50 μM, [e]nd, not determined, [f]–, not tested, [g] positive control for NO, [h] positive control for NF-κB.

triplet methyl group at δ_H 0.93 (t, J = 7.0 Hz)/δ_C 13.0 (C-7), an oxygenated methine group at δ_H 3.83 (1H, m)/δ_C 76.3 (C-2), and four methylene group signals at [δ_H 1.63 - 1.43 (2H, m)/δ_C 36.3 (C-3), 1.37 (2H, m)/δ_C 24.6 (C-4), 1.37 (2H, m)/δ_C 22.2 (C-4), 1.33 (2H, m)/δ_C 31.7 (C-4)], indicating the presence of a heptan-2-oyl moiety, confirmed by two- and three-bond ^{1}H-^{13}C HMBC and ^{1}H-^{1}H COSY correlations (Fig. 2). The NMR spectra revealed two additional anomeric proton signals at δ_H 4.33 (d, J = 8.0 Hz, H-1') and at δ_H 4.35 (d, J = 7.6 Hz, H-1"), along with 12 carbons for the inner β-D-glucose moiety at δ_C 102.6, 73.4, 76.2, 70.0, 75.4, and 68.3 and for the terminal β-D-xylofuranose group at δ_C 104.0, 73.8, 76.5, 69.7, and 65.5.[16] The HMBC correlation from the anomeric proton (H-1) of xylofuranose to the downfield shifted oxymethylene carbon (C-6) of glucose supported the connectivity of two sugar unites as the β-D-xylofuranosyl-(1 → 6)-β-D-glucopyranoside (Fig. 2).[16] The linkage of the heptan-2-oyl and the disaccharide moieties was established on the basis of the HMBC correlation between H-1' and C-2 (Fig. 2). Thus, compound **1** was identified as the alkylated disaccharide, heptanyl 2-*O*-β-D-xylofuranosyl-(1 → 6)-β-D-glucopyranoside.[16]

Compound **2** was obtained as white amorphous powder. The molecular formula was evaluated as *m/z*237 $[M + H]^+$ by the positive LCMS mass spectrometry. The IR spectrum showed one or more hydroxy group(s) and an alkyl group at 3345 and 2842 cm^{-1}, respectively. The NMR spectra of **2** showed similar patterns compare to that of **1**. However, **2** displayed more simple NMR signals: an oxygenated methylene group at δ_H 3.53 (1H, m)/δ_C 60.2 (C-1), a triplet methyl group at δ_H 0.96 (t, J = 7.4 Hz)/δ_C 13.0 (C-4), and two methylene groups at [δ_H 1.58 (2H, m)/δ_C 31.9 (C-2), 1.42 (2H, m)/δ_C 19.0 (C-3)], indicating the presence of a *n*-butanoyl group, and an additional anomeric proton signal at δ_H 3.75 (d, J = 8.0 Hz, H-1'), along with 6 carbon signals at δ_C 100.2, 69.2, 69.6, 63.7, 70.1, and 62.0, indicating the β-D-glucose.[17] The linkage of the two moieties was established on the basis of the HMBC correlations of H-1'/C-1 and H-1/C-1'. Accordingly, compound **2** was identified as *n*-butyl-β-D-glucopyranoside.[17]

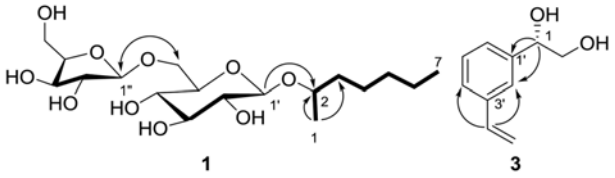

Fig. 2. Important HMBC (arrow) and COSY (bold) correlations for **1** and **3**.

Compound **3**, $[\alpha]_D^{25}$ +32 (*c* 0.12, MeOH), was obtained as colorless oil. The LCMS gave the molecular ion peak at *m/z*165 $[M + H]^+$.The NMR and HSQC spectra displayed an oxygenated methine group at δ_H 4.70 (1H, dd, J = 7.2, 4.8 Hz)/δ_C 73.0 (C-1), an oxymethylene group at δ_H 3.65 (2H, m)/δ_C 66.1 (C-2), indicating an ethane-diol molecule, a mono-substituted *exo*-methylene group at δ_H 6.77 (1H, dd, J = 18.0, 10.8 Hz)/δ_C 135.1 (C-7') and at δ_H 5.81 (1H, dd, J = 17.6, 10.8 Hz, H-8'a) & 5.25 (1H, dd, J = 10.8, 0.8 Hz, H-8'b)/δ_C 111.9 (C-8'), four protonated aromatics at δ_H 7.28 - 7.37 (3H, m, H-4' to H-6')/δ_C 124.0-124.5 (C-4' to C-6') and at δ_H 7.42 (1H, br s)/δ_C 122.5 (C-2'), and two quaternary aromatic carbons at δ_C 143.0 (C-1') and 140.0 (C-3'), indicating an ethenyl-phenyl structure. The HMBC correlations from H-1 to C-1'/C-2' and from H-7 to C-2'/C4' supported the structure of **3** as (3-ethenyl-phenyl)-1,2-ethanediol. The relative configuration of C-1 in **3** was

determined as *S* form by comparison of its optical rotation with a published value, $[\alpha]_D^{16}$ +8.7 (Fig. 2).[18] On the basis of the above data, **3** was identified as (1*S*)-(3-ethenyl-phenyl)-1,2-ethanediol.[18]

Compound **4**, $[\alpha]_D^{25}$ −28 (*c* 0.12, MeOH), was obtained as a white amorphous powder. The NMR spectra of **4** revealed two ester carbonyl carbons, a methylene group, and an oxygenated methine group, assigned as 2-hydroxybutanedioic acid. The relative configuration of **4** was determined as *S* form by comparison of its optical rotation with a published value, $[\alpha]_D^{24}$ −31.[19] Accordingly, **4** was confirmed as (2*S*)-2-hydroxybutanedioic acid.

The remaining compound **5** was identified as daucosterol by comparison of its NMR data with a published value.[20]

To the best of our knowledge, compounds **1** – **3** were isolated for the first time from this plant source. In particular, heptanyl 2-*O*-*β*-D-xylofuranosyl-(1→6)-*β*-D-glucopyranoside **1** and phenyl ethanediol **3** were only reported from *Bidens pilose*[16] and the culture broth of *Boletus edulis*, respectively.[18]

The isolated compounds were evaluated for their cancer chemopreventive potential based on their ability to inhibit nitric oxide (NO) production and tumor necrosis factor alpha (TNF-α)-induced NF-κB activity, and quinonone reductase-1 (QR1)-inducing effect. Among the isolates, compounds **2** and **3** showed moderate quinone reductase-1 (QR-1) inducing activities with IR values of 1.4 and 1.8, respectively, whereas the other isolates including **2** and **3** showed weak or no inhibitory activities against the TNF-α-induced NF-κB and NO production. Although the biological activities of **2** and **3** displayed moderate action on QR-1 inducing activity *in vitro* assay system, alkylated phenyls and their synthetic products, and alkyl ester glycoside derivatives have potential anti-oxidant and anti-cancer activities[21,22] and inhibitory effect against TPA-induced Epstein-Barr virus early antigen (EBV-EA) activation in *Raji* cells,[23] respectively. Thus, more studies are required to further biological actions.

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