New Anthraquinone and Iridoid from the Fruits of *Morinda citrifolia*

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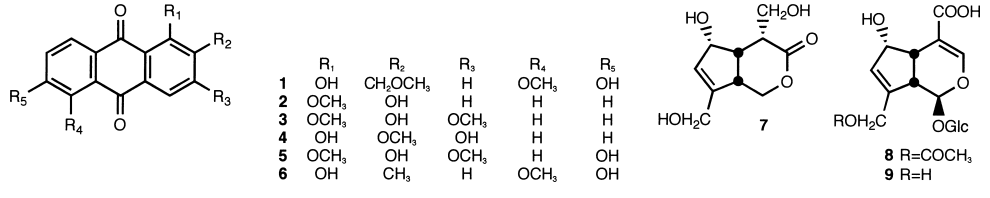
From the fruits of *Morinda citrifolia* L., one new anthraquinone, 5,15-*O*-dimethylmorindol, together with five known anthraquinones and one new iridoid, morindacin, together with two known iridoids, were isolated. Their structures were elucidated by analysis of spectroscopic data.

Key words *Morinda citrifolia*; Rubiaceae; anthraquinone; iridoid

Morinda citrifolia L. (Rubiaceae), known as "noni", is a small tree that grows widely across Polynesia. The roots, barks, stems, leaves and fruits have been used traditionally as a folk medicine for the treatment of many diseases[1] including diabetes, hypertension[2] and cancer.[3] Furthermore, today "noni juice", which is made from the fruits of this plant, is widely drunk for the purported prevention of lifestyle-related diseases such as diabetes, hypertension, cardiopathy and cerebral apoplexy caused by arteriosclerosis. In our previous study of the bioactive constituents of *M. citrifolia* fruits for the prevention of arteriosclerosis, six lignans were isolated as active components.[4] The rubiaceous plant is well-known for its anthraquinone and iridoid constituents. In earlier studies of anthraquinones from *M. citrifolia*, morenone-1, morenone-2, nordamnacanthal, morindone, rubiadin, rubiadin-1-methyl ether and 7-hydroxy-8-methoxy-2-methylanthraquinone were identified in the root.[5—7] Additionally, morindone, physcion and physcion-8-*O*-α-L-arabinopyranosyl(1—3)-[β-D-galactopyranosyl(1—6)]-β-D-galactopyranoside were found in its heartwood,[8] while 6,8-dimethoxy-3-methylanthraquinone 1-*O*-β-rhamnopyranosyl(1—4) β-D-glucopyranoside has been isolated from the flower.[9] In studies of iridoids from *M. citrifolia*, asperuloside,[10] asperulosidic acid,[11] 6α-hydroxyadoxoside,[12] 6β,7β-epoxy-8-epi-splendoside,[12] borreriagenin,[12] citrifolinin B,[12] deacetylasperuloside[12] and dehydromethoxygaertneroside[12] were all isolated from the fruits, while citrifolinin A and citrifolinoside were isolated from the leaves.[13,14] The present paper describes the isolation and characterization of one new anthraquinone, 5,15-dimethylmorindol (**1**), together with five known anthraquinones, alizarin-1-methylether (**2**),[15] anthragallol-1,3-dimethylether (**3**),[16] anthragallol-2-methylether (**4**),[17] 6-hydroxy-anthragallol-1,3-dimethylether (**5**)[18] and morindone-5-methylether (**6**),[19] and one new iridoid, morindacin (**7**), together with two known iridoids, asuperlosidic acid (**8**)[20] and deacetylasperulosidic acid (**9**).[20]

Compound **1** was obtained as a yellow amorphous powder. The molecular formula of **1** was determined by HR-EI-MS to be $C_{17}H_{14}O_6$. The ^{13}C-NMR spectrum indicated 17 carbon signals, including two methoxy carbons (δ 58.89, 62.36), one methylene carbon (δ 68.56) and two carbonyl carbons (δ 187.78, 181.91). In the ^{1}H-NMR spectrum, two pairs of *ortho*-coupled proton signals [one at δ 7.78 and 7.73 (each 1H, d, J=7.8 Hz), and the other at δ 8.14 and 7.35 (each 1H, d, J=8.5 Hz)] were observed. Furthermore, the presence of two methoxy groups, one methylene group and a hydrogen-bonded hydroxy group were suggested from the ^{1}H resonances of δ 4.03 and 3.51 (each 3H, s), 4.64 (2H, s) and 13.01 (1H, s). The regiochemistry of each functional group was determined by a HMBC experiment (Fig. 2). From the above results, compound **1** was characterized as 1,6-dihydroxy-5-methoxy-2-methoxymethylanthraquinone. This compound was named as 5,15-*O*-dimethylmorindol, as it is the 5,15-dimethylether of morindone 15-alcohol.

Compound **7**, a colorless syrup, gave a molecular formula of $C_{10}H_{14}O_5$ by HR-FAB-MS. The IR spectrum of **7** indicated absorption bands due to hydroxyl and lactone functions at 3400 cm^{-1} and 1743 cm^{-1}, respectively. The ^{1}H- and ^{13}C-NMR spectra of **7** showed signals assignable to a trisubstituted double bond [δ_H 5.84 (H-7), δ_C 125.07, 153.35 (C-7, 8)], three oxygen-bearing methylenes [δ_H 3.72, 3.79 (H-1), δ_C 60.79 (C-1)], [δ_H 4.16, 4.22 (H-10), δ_C 60.54 (C-10)] and [δ_H 3.84, 3.90 (H-11), δ_C 62.82 (C-11)] and one oxygen-bearing methine [δ_H 5.40 (H-6), δ_C 88.27 (C-6)]. The connectivities of the quaternary carbons (C-3, C-8) were deduced by a HMBC experiment (Fig. 3). In the HMBC spectrum, one quaternary carbon signal at δ 180.93 (C-3) was correlated with methine signals at δ 2.96 (H-4) and 3.33 (H-5), and the other quaternary carbon signal at δ 153.35 (C-8) was correlated with methylene signals at δ 3.72, 3.79 (H-1) and 4.16, 4.22 (H-10). Acetylation of **7** with acetic anhydride in pyridine yielded the triacetate (**7a**), suggesting that **7** possessed three hydroxyl groups. From the above evidence and with the aid of ^{1}H–^{1}H and ^{13}C–^{1}H COSY spectra, the planar structure was established. Furthermore, the relative stereostructure of **7** was characterized on the basis of a NOESY



	R_1	R_2	R_3	R_4	R_5
1	OH	CH_2OCH_3	H	OCH_3	OH
2	OCH_3	OH	H	H	H
3	OCH_3	OH	OCH_3	H	H
4	OH	OCH_3	OH	H	H
5	OCH_3	OH	OCH_3	H	OH
6	OH	CH_3	H	OCH_3	OH

Fig. 1. Anthraquinones and Iridoids from the Fruits of *M. citrifolia*

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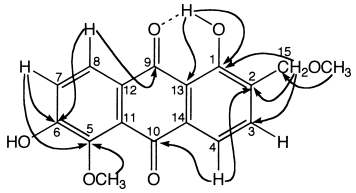

Fig. 2. HMBC (Arrows) Correlations of **1**

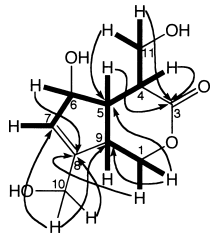

Fig. 3. ^{1}H–^{1}H COSY (Bold Lines) and HMBC (Arrows) Correlations of **7**

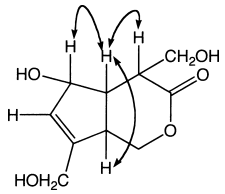

Fig. 4. NOESY (Arrows) Correlations of **7**

experiment (Fig. 4): *i.e.*, the proton signal at δ 3.33 (H-5) showed a NOESY correlation with proton signals at δ 2.96 (H-4), 5.40 (H-6) and 3.10 (H-9), indicating that the stereochemical relationship of a hydroxymethyl group at C-4 and the hydroxy group at C-6 was *syn*. Therefore, the structure of **7** was determined to be 1,4-bis(hydroxymethyl)-3-hydroxy-3,4,6,7,3a,7a-hexahydro-6-oxainden-5-one (IUPAC nomenclature). This is the first report of this compound from a natural source, and it has been named morindacin.

Experimental

General Procedures and Plant Material General experimental procedures and plant material have been described in an earlier publication.[4]

Extraction and Isolation The extraction and partition processes have been published earlier.[4] The methanol extract (89 g) of *M. citrifolia* fruits was dissolved in a MeOH–H_2O mixture (1 : 3, 1 l) and sequentially partitioned with $CHCl_3$, EtOAc and *n*-BuOH (each 1 l×3 times). The $CHCl_3$ soluble phase (44 g) was chromatographed on Sephadex LH-20 using $CHCl_3$–MeOH (1 : 1) to give an anthraquinone-containing fraction. This fraction was subjected repeatedly to SiO_2 column chromatography using a hexane–EtOAc solvent system to afford compounds **1** (18.2 mg), **2** (6.0 mg), **3** (8.3 mg), **4** (8.0 mg), **5** (1.5 mg) and **6** (6.3 mg). The *n*-BuOH soluble phase (9.7 g) was chromatographed on Sephadex LH-20 using MeOH to give an iridoid-containing fraction. This fraction was subjected repeatedly to Rp-18 column chromatography using MeOH–H_2O (1 : 4), and to SiO_2 column chromatography using $CHCl_3$–MeOH (4 : 1) to afford compounds **7** (29.2 mg), **8** (36.9 mg) and **9** (119.3 mg).

5,15-Dimethylmorindol (**1**): Yellow amorphous powder; HR-EI-MS: *m/z* $[M]^+$ 314.0797 (Calcd for $C_{17}H_{14}O_6$: 314.0790); IR ν_{max}^{KBr} cm^{-1}: 3400, 1668, 1632, 1566, 1508, 1429, 1362, 1279, 1258, 1117, 961; UV λ_{max}^{MeOH} nm (log ε): 412 (3.86), 267 (4.25), 224 (4.45), 203 (4.17); ^{1}H-NMR (400 MHz) and ^{13}C-NMR (100 MHz): see Table 1.

Morindacin (**7**): Colorless syrup; $[\alpha]_D^{26}$ +2.0° (*c*=0.2, MeOH); HR-FAB-MS: $[M+H]^+$ *m/z* 215.05091 (Calcd for $C_{10}H_{15}O_5$: 215.0919); IR ν_{max}^{KBr} cm^{-1}: 3400, 1743, 1635, 1386, 1190, 1051; ^{1}H-NMR (400 MHz) and ^{13}C-NMR (100 MHz): see Table 2.

Table 1. The ^{1}H- and ^{13}C-NMR Spectral Data for **1** in $CDCl_3$

No.	^{13}C	^{1}H
1	159.48	
2	133.94	
3	134.46	7.73 (d, 7.8)
4	118.98	7.78 (d, 7.8)
5	146.89	
6	156.07	
7	120.09	7.35 (d, 8.5)
8	125.57	8.14 (d, 8.5)
9	187.78	
10	181.91	
11	125.81	
12	126.96	
13	114.89	
14	133.27	
15	68.56	4.64 (s)
5-OMe	62.36	4.03 (s)
15-OMe	58.89	3.51 (s)

Coupling patterns and coupling constants (*J*) in Hz are given in parentheses.

Table 2. The ^{1}H- and ^{13}C-NMR Spectral Data for **7** in CD_3OD

No.	^{13}C	^{1}H
1	60.79	3.79 (dd, 11.4, 4.5)
		3.72 (dd, 11.4, 6.7)
3	180.93	
4	45.89	2.96 (ddd, 6.0, 4.7, 3.7)
5	43.97	3.33 (dt, 7.7, 6.0)
6	88.27	5.40 (br d, 7.7)
7	125.07	5.84 (quint-like)
8	153.35	
9	49.99	3.10 (m)
10	60.54	4.22 (ddd, 15.0, 2.4, 1.2)
		4.16 (ddd, 15.0, 2.8, 1.7)
11	62.82	3.90 (dd, 10.8, 4.7)
		3.84 (dd, 10.8, 3.7)

Coupling patterns and coupling constants (*J*) in Hz are given in parentheses.

Acetylation of 7 Compound **7** (2.0 mg) was acetylated with Ac_2O-pyridine, and the product was purified by column chromatography on SiO_2 using hexane–EtOAc (5 : 1) to yield triacetate **7a** (1.1 mg). **7a**: Colorless amorphous powder; ^{1}H-NMR (400 MHz, CD_3OD) δ: 5.95 (1H, d, *J*=1.9 Hz, H-7), 5.42 (1H, m, H-6), 4.72 (2H, m, H-10), 4.42 (1H, dd, *J*=11.9, 4.9 Hz, H-1a), 4.40 (1H, dd, *J*=11.1, 4.0 Hz, H-11a), 4.32 (1H, dd, *J*=11.1, 4.3 Hz, H-11b), 4.21 (1H, dd, *J*=11.9, 3.4 Hz, H-1b), 3.27 (2H, m, H-5, 9), 3.03 (1H, m, H-4), 2.08, 2.07, 1.98 (each 3H, s, $COCH_3$).

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