JOURNAL OF NATURAL PRODUCTS

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An Iridoid Glucoside and the Related Aglycones from *Cornus florida*

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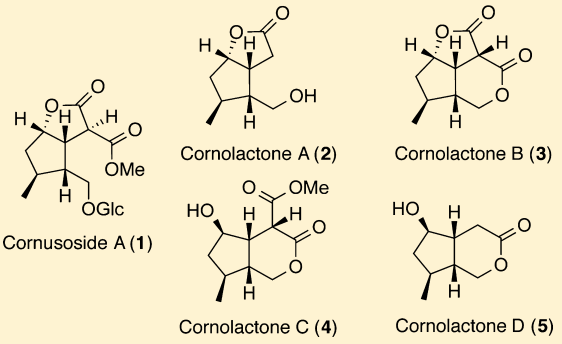
Supporting Information

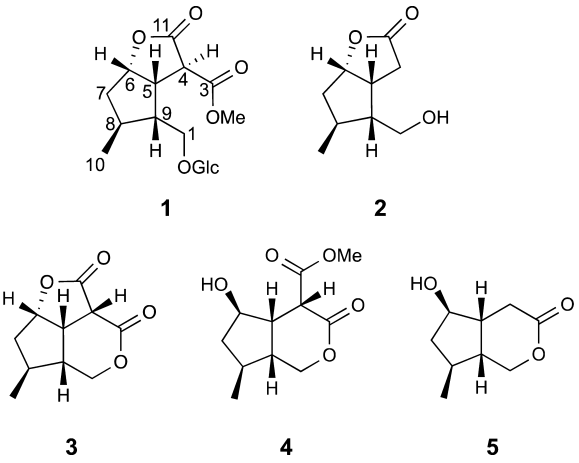
ABSTRACT: A new iridoid glucoside, cornusoside A (**1**), and four new natural product iridoid aglycones, cornolactones A–D (**2**–**5**), together with 10 known compounds were isolated from the leaves of *Cornus florida*. The structures of compounds **1**–**5** were established by interpretation of their spectroscopic data. Cornolactone B (**3**) is the first natural *cis*-fused tricyclic dilactone iridoid containing both a five- and a six-membered lactone ring. A biosynthesis pathway is proposed for cornolactones C (**4**) and D (**5**), the C-6 epimers of compounds **1**–**3**.



The plant genus *Cornus* (dogwood) belongs to the family Cornaceae and consists of approximately 55 species distributed mainly in the northern hemisphere, eastern Asia, and eastern and northern parts of the United States.[1] This genus is a rich source of diverse iridoid glucosides, which have raised interest because of their wide range of promising bioactivities. These include antidiabetic,[2,3] antioxidant,[4] anti-inflammatory,[5] antiamnesic,[6] and immunesuppressive effects.[7]

Cornus florida L., commonly known as flowering dogwood, is a tree native to eastern North America that has been traditionally used for the treatment of malaria.[8,9] Previous chemical investigations of *C. florida* have resulted in the isolation of a number of compounds including anthocyanins and other flavonoids, triterpenoids, and sterols.[9,10] The anthocyanins impart bright colors to several fruits and vegetables and possess anti-inflammatory,[10−12] antioxidative,[11,12] antineoplastic,[10,13] and antidiabetic activities.[14]

In the present study the chemical constituents of *C. florida* collected from Oxford, Mississippi, were investigated. A large-scale extraction of the leaves of *C. florida* yielded a new iridoid glucoside, cornusoside A (**1**), and four new natural product iridoid aglycones, cornolactones A–D (**2**–**5**). Cornolactone A (**2**) was previously reported as a synthetic intermediate in the enantioselective synthesis of semperoside A;[15] however, this is the first report of this compound from a natural source. In addition, 10 known compounds were also isolated, which included five iridoids, two megastigmane compounds, and two ellagic acid derivatives, together with a flavonoid. The structures of the new compounds were assigned by detailed spectroscopic analysis and those of the known compounds by comparison with literature data. Cornusoside A (**1**) is one of a small number of C_{10} iridoid glucosides with a ring-opening between C-1 and O-2 and a γ-lactone linkage between C-6 and C-11. Cornolactone B (**3**) is the first natural *cis*-fused tricyclic dilactone iridoid containing both a five- and a six-membered lactone ring. Herein are reported the isolation and structure elucidation of **1**–**5** and a possible biosynthetic pathway to cornolactones C (**4**) and D (**5**) as C-6 epimers of compounds **1**–**3**.



A 90% aqueous ethanol extract of the dried leaves of *C. florida* (15 kg) was fractionated initially on silica gel (step gradient elution of hexane to EtOAc to MeOH). The 20% MeOH in EtOAc was then subjected to column chromatography on polymeric HP-20 (step gradient elution 10% Me_2CO

Received: March 12, 2014
Published: August 21, 2014

dx.doi.org/10.1021/np5002362 | *J. Nat. Prod.* 2014, 77, 2138−2143

in H_2O to 100% Me_2CO). The 20% Me_2CO fraction was then subjected to repeated fractionation on either polymeric HP-20ss, reversed-phase C_{18}, normal-phase silica gel, or molecular exclusion Sephadex LH-20 column chromatography followed by a series of HPLC separations on either a PRP-1 column or a C_8 or C_{18} column to yield cornusoside A (**1**), cornolactones A–D (**2**–**5**), and the known compounds alternosides A,[16] hastatoside,[17] cornin,[17] dihydrocornin,[18] cornalternoside,[16] lauroside A,[19] (5*S**,6*R**)-9-hydroxymegastigm-7-en-3-one,[20] 3,3′-dimethyl-4-*O*-*β*-D-glucopyranosylellagic acid,[21] 3,4,3′-trimethyl-4′-*O*-*β*-D-glucopyranosylellagic acid,[22] and isoquercitrin.[23]

Cornusoside A (**1**) was isolated as a colorless gum. Its molecular formula, $C_{17}H_{26}O_{10}$, determined from the HRESIMS of the $[M + Na]^+$ at *m*/*z* 413.1417, required five degrees of unsaturation. A preliminary analysis of the ^1H and ^{13}C NMR data (Table 1) revealed two ester carbonyl groups (δ_C 172.8 and 169.0; IR 1730 cm^{-1}), an oxygenated methine [δ_H 5.03 (1H, t, *J* = 6.8 Hz, H-6); δ_C 83.6 (C-6)], an oxymethylene [δ_H 3.98 (1H, dd, *J* = 10.4, 3.6 Hz, H-1a), δ_H 3.38 (1H, m, H-1b); δ_C 66.7 (C-1)], a methoxy [δ_H 3.68 (3H, s); δ_C 52.7], and signals that could be attributed to a glucopyranosyl group. The coupling constant of the anomeric proton (δ_H 4.05, 1H, d, *J* = 7.8 Hz) suggested a *β*-configuration of the glucose. The presence of the *β*-D-glucopyranosyl group was confirmed by acid hydrolysis. These data led to the preliminary conclusion that **1** is an iridoid glucoside and accounted for three of the five double-bond equivalents, indicating the compound to be bicyclic.

The terpenoid portion of **1** was determined by detailed analysis of its NMR data (Figure 1). The observation of ^1H–^1H COSY correlations from H-6 to H-5 and H-7, H-7 to H-8, H-8 to H-9 and the methyl doublet H_3-10, and H-9 to H-5 established the presence of a cyclopentane ring with methyl substitution at C-8. An additional COSY correlation observed between H-9 and H-1a and H-1b of the oxygenated methylene group at C-1 together with an HMBC correlation from H-1′ to C-1 and from H-1 to C-1′ confirmed the connection of the *β*-D-glucopyranosyl group to C-1 and the connection of C-9 to C-1. Remaining to be assigned were two ester carbonyl carbons, C-3 (δ_C 169.0) and C-11 (δ_C 172.8), a methine, H-4 (δ_H 3.94; δ_C 47.8), and a methoxy group (δ_H 3.68; δ_C 52.7). HMBC correlations observed between H-6 (δ_H 5.03) and the ester carbonyl carbon at δ_C 172.8 (C-11) and from H-5 (δ_H 3.37) to both C-4 and C-11 established the presence of a *γ*-lactone ring. A COSY correlation observed between H-5 and H-4 and an HMBC correlation from H-4 to C-11 further supported this assignment. Finally, HMBC correlations observed from H-4, H-5, and the methoxy signal at δ_H 3.68 (OMe-3) to the remaining ester carbonyl carbon at δ_C 169.0 (C-3) established the connection of C-4 to C-3 and the presence of a methyl ester at C-3.

The relative configuration of **1** was determined by NOE correlations observed in a NOESY experiment and by scalar coupling (Figure 2). NOE correlations from H-5 to H-6 and H-9 together with correlations from H-7*β* to H-6 and H_3-10 indicated that H-5, H-6, H-9, and Me-10 are on the same side

Table 1. NMR Spectroscopic Data for Cornusoside A (1) and Cornolactone A (2)[a]

	1[b]		**2**[c]	
position	δ_C, mult.	δ_H (*J* in Hz)	δ_C, mult.	δ_H (*J* in Hz)
1a	66.7, CH_2	3.98, dd (10.4, 3.6)	61.4, CH_2	3.87, dd (11.0, 4.0)
1b		3.38, m		3.58, dd (11.0, 9.0)
3	169.0, qC			
4*α*	47.8, CH	3.94, d (5.6)	29.7, CH_2	2.66, dd (18.8, 4.7)
4*β*				2.59, dd (18.8, 9.8)
5	45.5, CH	3.37, q (7.2)	40.2, CH	3.14, m
6	83.6, CH	5.03, t (6.8)	84.9, CH	5.00, t (6.0)
7*α*	40.6, CH_2	1.99, dd (13.6, 6.0)	41.9, CH_2	2.20, dd (14.0, 5.0)
7*β*		1.47, ddd (13.6, 11.6, 6.0)		1.44, ddd (14.0, 11.8, 5.5)
8	32.0, CH	1.90, m	32.8, CH	1.82, m
9	48.0, CH	1.83, m	50.6, CH	1.79, m
10	17.3, CH_3	0.95, d (5.6)	17.6, CH_3	1.02, d (5.6)
11	172.8, qC		178.3, qC	
OMe	52.7, CH_3	3.68, s		
1′	103.0, CH	4.05, d (7.8)		
2′	73.3, CH	2.92, td (8.0, 4.4)		
3′	76.7, CH	3.10, m		
4′	70.1, CH	3.03, m		
5′	76.9, CH	3.07, d (3.9)		
6a′	61.1, CH	3.64, m		
6b′		3.43, m		
OH-2′		4.72, d (4.3)		
OH-3′		4.94, d (5.0)		
OH-4′		4.93, d (5.5)		
OH-6′		4.45, t (5.9)		

[a] ^1H NMR measured at 400 MHz, ^{13}C NMR measured at 100 MHz. [b] Measured in DMSO-d_6. [c] Measured in $CDCl_3$.

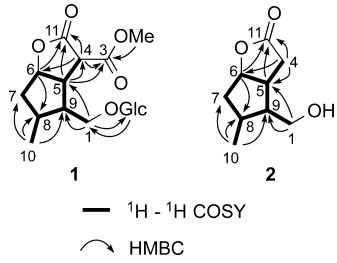


Figure 1. Selected 2D NMR correlations for cornusoside A (**1**), and cornolactone A (**2**).

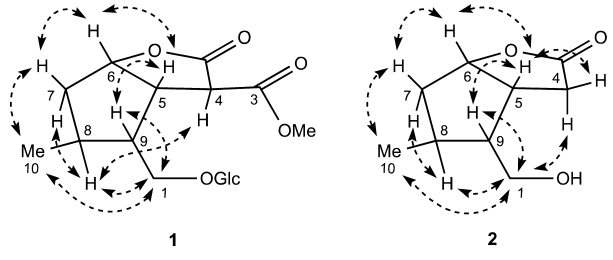


Figure 2. Selected NOE correlations observed for cornusoside A (**1**) and cornolactone A (**2**).

dx.doi.org/10.1021/np5002362 | J. Nat. Prod. 2014, 77, 2138–2143

of the cyclopentane ring in a β-orientation. NOE correlations from H-8 to H-7α and H-1 confirmed the α-orientation of H-8 and the glucosylated side chain. A long-range *W*-coupling in the COSY spectrum between Me-10 and H-7α was consistent with the 1,2-diaxial arrangement of these two groups. Finally, the α-orientation of H-4 was indicated by a NOE correlation observed between H-4 and H-8. This was substantiated by a small coupling (5.6 Hz) observed between H-4 and H-5 that was similar to the coupling constants (ca. 5.0 Hz in both cases) of the previously reported C-1 to O-2 ring-opened iridoids gelsemiol[24] and borreriagenin.[25] The absolute configuration of **1** was determined based on biogenetic grounds in that nearly all iridoids found in Nature have a configuration of 5*S* and 9*R* and by analogy to the known co-isolated compounds that were found to have closely comparable NMR data and similar optical rotation values. Thus, the configuration of cornusoside A (**1**) was defined as 4*S*,5*S*,6*S*,8*S*,9*R*. Additional support for the absolute stereochemistry came from the isolation of cornolactone A (**2**), previously reported as a synthetic intermediate in the total synthesis of the iridoid semperoside A.[15] Since this is the first report of **2** from a natural source, the isolation, structure elucidation, and full spectroscopic data are reported.

Cornolactone A (**2**), isolated as a colorless gum, showed a $[M + H]^+$ ion at *m*/*z* 171.1010 in the HRESIMS corresponding to the molecular formula $C_9H_{14}O_3$, requiring four degrees of unsaturation. An initial inspection of the NMR data revealed **2** to be similar to **1**, except for the absence of the signals for the methine at C-4, the methyl ester at C-3, and the glucopyranosyl group and the appearance of signals for a diastereotopic methylene at δ_H 2.66 (1H, dd, *J* = 18.8, 4.7 Hz, H-4α) and δ_H 2.59 (1H, dd, *J* = 18.8, 9.8 Hz, H-4β). This suggested that **2** is an iridoid aglycone missing a C-3 methyl ester moiety. The observation of ^{1}H−^{1}H COSY correlations from both H_2-4 to H-5 and HMBC correlations from H_2-4 to C-5, C-6, and C-11 and from H-6 to C-11 further supported this assignment (Figure 1). The similarity of proton−proton coupling constants and ^{1}H and ^{13}C NMR chemical shifts together with the NOESY spectrum of **2** showed the same relative configuration as **1** at the four chiral centers (Figure 2). Additional NOE correlations from H-4α to H-1b of the oxygenated methylene and from H-4β to H-5 confirmed this assignment. Compound **2** was found to have near-identical NMR data and a comparable optical rotation value ($[\alpha]^{18}_D$ +3.2) (lit. $[\alpha]_D$ +14.7) to that of the reported synthetic compound.[15]

Cornolactone B (**3**) was isolated as a colorless gum. The molecular formula of cornolactone B (**3**), $C_{10}H_{12}O_4$, as determined from the HRESIMS of the $[M + H]^+$ ion at *m*/*z* 197.0811, required five degrees of unsaturation. Analysis of the NMR data (Figure 3) revealed **3** to be similar to **2** except for the absence of the signals for the methylene group at C-4 and the addition of signals for an ester carbonyl carbon at δ_C 164.7 (C-3) and a methine δ_H 3.78 (1H, d, *J* = 9.6 Hz, H-4). Having accounted for all protons, carbons, and oxygens in the molecule and four of the five double-bond equivalents, as required by the molecular formula, this indicated that cornolactone B (**3**) is tricyclic. The only possible connection was between the C-1 oxygen and the ester carbonyl carbon C-3 to form a δ-lactone unit. The connection was confirmed with HMBC correlations observed between both H_2-1 (δ_H 4.47, 4.05) and H-4 (δ_H 3.80) to the ester carbonyl carbon at δ_C 170.2 (C-3). The relative configuration of **3** at C-5, C-6, C-8, and C-9 was determined to be identical to that of **1** and **2** by NOE correlations observed in a NOESY experiment and scalar coupling (Figure 4). The absence of any NOE correlations observed to or from H-4 made it difficult to assign the configuration at C-4. However, the presence of a large ^{1}H NMR coupling constant (*J* = 9.6 Hz) between H-4 and H-5 suggested the *cis*-relationship of these two protons and the β-orientation of H-4. This coupling constant is consistent with that observed for the *cis*-fused tricyclic iridoids semperoside (*J* = 10.5 Hz), 9-hydroxysemperoside (*J* = 11.4 Hz), and dihydrobrasoside (*J* = 10.5 Hz),[24] together with the dilactone compounds, asperuloside tetraacetate lactone (*J* = 9.8 Hz) and dihydroasperuloside tetraacetate lactone (*J* = 10.0 Hz), produced from the oxidation of the iridoid glucoside asperuloside.[26] Thus, the configuration of cornolactone B (**3**) was defined as 4*S*,5*S*,6*S*,8*S*,9*R*.

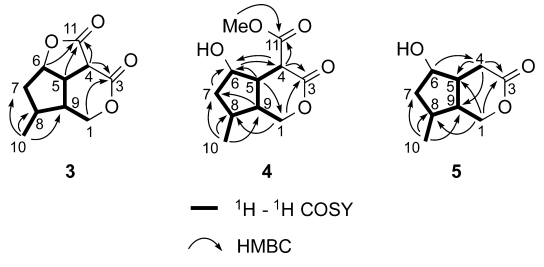


Figure 3. Selected 2D NMR correlations for cornolactones B−D (**3**−**5**).

Cornolactone C (**4**) was isolated as a colorless gum. The molecular formula of this compound, $C_{11}H_{16}O_5$, as determined from the HRESIMS of the $[M + Na]^+$ ion at *m*/*z* 251.0884, required four degrees of unsaturation. The NMR data of **4** were similar to those of cornolactone B (**3**), except that the addition of a methoxy group (δ_H 3.80; δ_C 53.4) was shown and H-6 [δ_H 5.04, t (5.5)] was shifted upfield by 1.22 ppm (Table 2) as compared to that of **3**. This suggested **4** is the product from methanolysis of the γ-lactone ring in **3**. An HMBC correlation from the methoxy signal at δ_H 3.80 (OMe-11) to δ_C 169.2 (C-11) confirmed the presence of a methyl ester at C-11. Additional ^{1}H−^{1}H COSY and HMBC correlations (Figure 3) further supported this assignment. The relative configuration of **4** was determined by NOE correlations observed in a NOESY experiment and scalar coupling (Figure 4). NOE correlations from H-5 to H-4, H-7β, and H-9, together with correlations from H_3-10 to H-7β and H-9, established the *cis*-fusion of the cyclopenta[*c*]pyran skeleton with H-4, H-5, H-7β, and H-9 in the β-orientation. Finally, the α-orientation of H-6 was indicated by an NOE correlation observed from H-6 to H-7α and H-8 on the underside of the cyclopentane ring. Thus, the configuration of cornolactone C (**4**) was defined as 4*R*,5*S*,6*R*,8*S*,9*R*.

Cornolactone D (**5**) was isolated as a colorless gum. The molecular formula, $C_9H_{14}O_3$, was determined from the HRESIMS of the $[M + H]^+$ ion at *m*/*z* 193.0834 (calcd 193.0835) and required three degrees of unsaturation. An initial inspection of the NMR data revealed that **5** is similar to **4**, except for the absence of the signals for the methine at C-4 and the methyl ester at C-3 and the appearance of a methylene group at δ_H 2.57 (2H, d, *J* = 5.2 Hz, H-4). This suggested that **5** did not have a C-11 methyl ester unit. The observation of ^{1}H−^{1}H COSY correlations from H_2-4 to H-5 and HMBC correlations from H_2-4 to C-3, C-5, and C-9, together with a correlation from H-6 to C-4, confirmed this assignment (Figure 3). The similarity of proton−proton coupling constants and ^{1}H and ^{13}C NMR chemical shifts together with a NOESY

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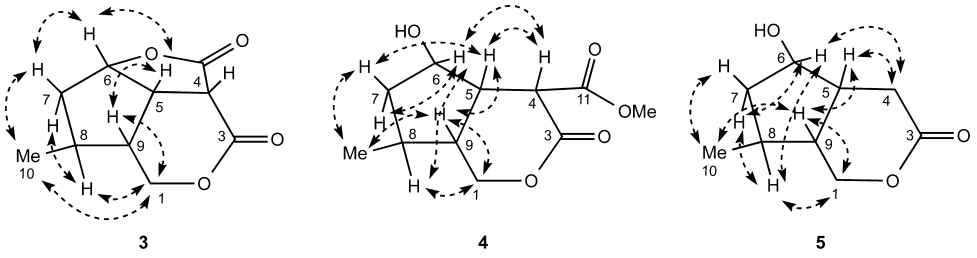

Figure 4. Selected NOE correlations observed for cornolactones B–D (**3**–**5**).

Table 2. NMR Spectroscopic Data for Cornolactones B–D (3–5) in $CDCl_3$[a]

	3		4		5	
position	δ_C, mult.	δ_H (J in Hz)	δ_C, mult.	δ_H (J in Hz)	δ_C, mult.	δ_H (J in Hz)
1a	68.8, CH_2	4.47, dd (12.2, 4.8)	69.3, CH_2	4.27, dd (12.0, 5.6)	68.6, CH_2	4.18, dd (11.6, 4.0)
1b		4.05, dd (12.2, 8.0)		4.00, dd (11.6, 6.0)		4.08, dd (12.0, 3.2)
3	164.7, qC		169.0, qC		174.0, qC	
4	45.7, CH	3.80, d (9.6)	50.4, CH	3.58, d (6.6)	32.4, CH_2	2.57, d (5.2)
5	42.3, CH	3.35, dt (9.5, 6.4)	46.1, CH	2.76, dt (11.6, 7.2)	42.6, CH	2.39, m
6	85.1, CH	5.04, t (5.5)	77.4, CH	3.82, ddd (10.2, 7.4, 6.0)	77.8, CH	3.71, ddd (10.2, 7.8, 5.5)
7α	41.2, CH_2	2.36, dd (14.8, 6.8)	43.2, CH_2	2.12, td (11.7, 6.0)	42.6, CH_2	2.01, m
7β		1.68, ddd (14.8, 9.4, 5.6)		1.38, dt (12.1, 10.2)		1.27, dt (12.1, 11.2)
8	34.2, CH	2.04, m	33.9, CH	1.78, m	33.1, CH	1.79, m
9	43.1, CH	2.10, m	42.8, CH	2.20, ddd (11.7, 9.4, 6.3)	43.2, CH	1.97, m
10	19.4, CH_3	1.12, d (6.7)	18.9, CH_3	1.07, d (6.3)	18.7, CH_3	1.03, d (6.4)
11	170.2, qC		169.2, qC			
OMe			53.4, CH_3	3.80, s		

[a] ^{1}H NMR measured at 400 MHz, ^{13}C NMR measured at 100 MHz.

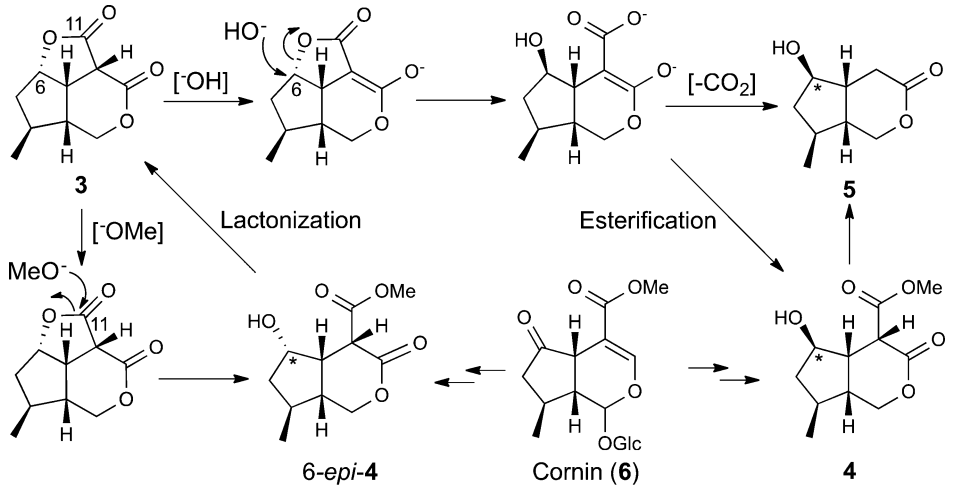

Figure 5. Plausible biosynthetic route to **3** and potential pathways to inversion of configuration at C-6 in **4** and **5**.

spectrum of **5** showed the same relative configuration as **4** at the four chiral centers (Figure 4). Thus, the configuration of cornolactone D (**5**) was defined as 5*R*,6*R*,8*S*,9*R*.

Compounds **1**–**5** and cornin were evaluated for cell growth inhibitory activities against human embryonic stem cells (BG02) and human breast cancer cell lines (MCF-7 and MDA-MB-231). No cytotoxicity was observed for any of the compounds at 100 μM except for slight cytotoxicty being observed for compound **5** against the MDA-MB-231 cell line. The compounds were also examined for agonistic activity against peroxisome proliferator-activated receptor γ (PPARγ), but no activity was observed.

Cornusoside A (**1**) is one of a small number of C_{10} iridoid glucosides where the δ-lactone is ring-opened between the C-1 and O-2 positions and contains a γ-lactone linkage between C-6 and C-11. Other examples reported include gelsemiol 3-*O*-β-D-glucoside,[24] gelsemiol 6′-*trans*-caffeoyl-1-glucoside,[27] and verbenabrasides A and B.[28] Cornolactone B (**3**) is the first natural *cis*-fused tricyclic dilactone iridoid containing both a five- and a six-membered lactone ring. Interestingly, cornolactones C (**4**) and D (**5**) have an opposite configuration at C-6 compared to that of compounds **1**–**3**. This suggests that rather than cleavage of the γ-lactone in **3** by methanolysis at C-11, to give the C-6 epimer of **4** (6-*epi*-**4**) with retention of configuration, an alternative biosynthetic pathway is necessary (Figure 5). A

dx.doi.org/10.1021/np5002362 | J. Nat. Prod. 2014, 77, 2138–2143

plausible pathway leading to inversion of configuration at C-6 could occur through the S_N2 hydrolysis of **3** at C-6, followed by esterification to give cornolactone C (**4**) or decarboxylation to give cornolactone D (**5**). Alternatively, cornolactones C (**4**) and D (**5**) could also have been formed from the reduction followed by oxidation of the co-isolated iridoid cornin (**6**). Previously it has been shown that reduction of **6** with $NaBH_4$ gives approximately a 1:1 mixture of both epimers at C-6.[18] It is also conceivable that 6-*epi*-**4** could undergo a lactonization reaction to give cornolactone B (**3**) and could provide an explanation of why no 6-*epi*-**4** was isolated.

EXPERIMENTAL SECTION

General Experimental Procedures. Optical rotations were measured on a JASCO P-2000 polarimeter (*c* g/100 mL) equipped with a halogen lamp (589 nm) and a 1 mL microcell. IR spectra were recorded on a Thermo Electronic Corporation Nicolet IR-100 spectrophotometer. All NMR spectra were acquired with a Varian MercuryPlus 400 spectrometer using solvent signals (DMSO-d_6: ^{1}H, δ_H 2.50; ^{13}C, δ_C 39.52; $CDCl_3$: ^{1}H, δ_H 7.24 ppm; ^{13}C, δ_C 77.23 ppm) as references. Short- and long-range ^{1}H−^{13}C correlations were determined with gradient-enhanced inverse-detected HSQC and HMBC experiments, respectively. NOE correlations were detected with NOESY experiments with a 0.5 s mixing time. The HRESIMS were obtained using an Agilent 6220 series TOF mass spectrometer. HPLC was performed on a Shimadzu LC-20AT instrument with a Shimadzu SPD-M20A UV/vis photodiode detector and a Shimadzu ELSD-LTII detector.

Plant Material. The leaves of *Cornus florida* L. were collected from a one-mile radius around Timber Lake, Oxford, Mississippi, during the spring and summer of 2011 by M.T.H. Voucher specimens are kept in the Hamann Laboratory at the University of Mississippi, School of Pharmacy, Oxford, MS (Cf2011).

Extraction and Purification Procedures. The leaves of *C. florida* (15.0 kg, dry weight) were extracted with 90% aqueous ethanol and dried under reduced pressure to give a crude extract (900 g). A portion of this crude extract (400 g) was separated on a silica gel column (20 × 70 cm) using a stepwise gradient of hexanes/EtOAc (100:0, 80:20, 50:50, and 0:100, v/v, each 3 L) and EtOAc/MeOH mixtures (80:20, 60:40, 50:50, and 0:100, v/v, each 3 L) to afford eight fractions. Fraction E (220 g) was then chromatographed on HP-20 (8 × 50 cm) using a stepwise gradient of acetone/water (10:90, 20:80, 50:50, 60:40, 80:20, and 100:0, v/v, each 2 L) to give six subfractions (Fr. E_1−E_6). Fraction E_2 (40 g) was chromatographed on a preparative C_{18} reversed-phase MPLC column (20 × 250 mm; 20−35% CH_3OH/H_2O over 40 min, 35−65% CH_3OH/H_2O over 20 min; flow rate: 12 mL/min) to afford eight subfractions (Fr. E_{2a}−E_{2h}). Fraction E_{2d} (4.0 g) was chromatographed on a preparative C_{18} reversed-phase HPLC column (Shim-pak RP-C_{18} column; 5 μm; 20 × 250 mm; 8−45% CH_3CN/H_2O over 120 min, 7 mL/min) to yield eight subfractions (Fr. $E_{2d\text{-}1}$−$E_{2d\text{-}8}$). Fraction $E_{2d\text{-}4}$ (50 mg) was purified by C_8 reversed-phase HPLC (Polar-C_8; 5 μm; 10 × 250 mm; 15−45% CH_3OH/H_2O over 90 min, 5 mL/min) to yield cornusoside A (**1**, 5.0 mg, t_R 76.2 min). Fraction $E_{2d\text{-}6}$ (832 mg) was subjected to silica gel column chromatography using CH_2Cl_2/MeOH (90:10 to 0:100, v/v) to afford nine fractions (Fr. $E_{2d\text{-}6\text{-}a}$−Fr. $E_{2d\text{-}h}$). Fraction $E_{2d\text{-}6\text{-}a}$ (320 mg) was further purified by C_8 reversed-phase HPLC (Polar-C_8; 5 μm; 10 × 250 mm; 5−25% CH_3CN/H_2O over 70 min, 5 mL/min) to yield two fractions. The first fraction (60 mg) was purified on a polymeric HPLC column (Hamilton PRP-1; 5 μm; 20 × 250 mm; 10−40% CH_3CN/H_2O over 50 min, 7 mL/min) to yield cornolactone C (**4**, 8.0 mg, t_R 41.5 min). The second fraction (40 mg) was chromatographed on a polymeric HPLC column (Hamilton PRP-1; 5 μm; 20 × 250 mm; 15−45% CH_3CN/H_2O over 70 min, 7 mL/min) to yield cornolactone B (**3**, 4.0 mg, t_R 37.9 min). Cornolactones A (**2**, 15.0 mg) and D (**5**, 12 mg) were purified from the remaining crude extract (500 g) using a similar procedure (see Supporting Information).

*Cornusoside A (**1**):* colorless gum; $[\alpha]^{18}_D$ −13.8 (*c* 0.01, MeOH); UV (MeOH) λ_{max} (log ε) 230 (3.4) nm; IR (KBr) ν_{max} 3355, 2922, 1730, 1073, 1020 cm^{-1}; ^{1}H and ^{13}C NMR (400 MHz, d_6-DMSO), see Table 1; HRESIMS *m*/*z* 413.1417 $[M + Na]^+$ (calcd for $C_{17}H_{26}O_{10}Na$, 413.1418).

*Cornolactone A (**2**):* colorless gum; $[\alpha]^{18}_D$ +3.3 (*c* 0.2, MeOH); UV (MeOH) λ_{max} (log ε) 237 (3.2) nm; IR (KBr) ν_{max} 3355, 2922, 1730, 1073, 1020 cm^{-1}; ^{1}H and ^{13}C NMR (400 MHz, $CDCl_3$), see Table 1; HRESIMS *m*/*z* 171.1010 $[M + H]^+$ (calcd for $C_9H_{15}O_3$, 171.1016).

*Cornolactone B (**3**):* colorless gum; $[\alpha]^{18}_D$ −7.5 (*c* 0.02, MeOH); UV (MeOH) λ_{max} nm 243 (3.1) nm; IR (KBr) ν_{max} 2944, 1658, 1023 cm^{-1}; ^{1}H and ^{13}C NMR (400 MHz, $CDCl_3$), see Table 2; HRESIMS *m*/*z* 197.0811 $[M + H]^+$ (calcd for $C_{10}H_{13}O_4$, 197.0808).

*Cornolactone C (**4**):* colorless gum; $[\alpha]^{18}_D$ +13.3 (*c* 0.03, MeOH); UV (MeOH) λ_{max} (log ε) 241 (3.1) nm; IR (KBr) ν_{max} 3337, 2952, 2937, 1733, 1650, 1017 cm^{-1}; ^{1}H and ^{13}C NMR (400 MHz, $CDCl_3$), see Table 2; HRESIMS *m*/*z* 215.0884 $[M + Na]^+$ (calcd for $C_{11}H_{16}O_5Na$, 215.0890).

*Cornolactone D (**5**):* colorless gum; $[\alpha]^{18}_D$ +18.5 (*c* 0.02, MeOH); UV (MeOH) λ_{max} (log ε) 244 (2.8) nm; IR (KBr) ν_{max} 3401, 2952, 1733, 1089, 1068 cm^{-1}; ^{1}H and ^{13}C NMR (400 MHz, $CDCl_3$), see Table 2; HRESIMS *m*/*z* 193.0834 $[M + Na]^+$ (calcd for $C_9H_{14}O_3Na$, 193.0835).

Acid Hydrolysis and Sugar Analysis. Cornusoside A (**1**) (2 mg) was hydrolyzed by using 1 M HCl (0.4 mL) at 100 °C for 2 h under argon and neutralized with Amberlite IR400. After drying under reduced pressure, the residue was dissolved in pyridine (0.4 mL) containing L-cysteine ethyl ester hydrochloride (2 mg) and heated at 60 °C for 1 h. A 0.4 mL solution of 3,5-dichlorophenyl isothiocyanate (2 mg) in pyridine was added to the mixture, which was heated at 60 °C for 1 h. The reaction mixture was directly analyzed by analytical HPLC on a Shim-pak RP-C_{18} column, 5 μm, 4.6 × 250 mm, by eluting with a gradient of 30−80% CH_3CN in H_2O with 0.02% HCOOH for 40 min and subsequent washing of the column with 100% CH_3CN at a flow rate 0.8 mL/min. In the acid hydrolysate of **1**, D-glucose was confirmed by comparison of the retention times of their derivatives with those of L-glucose and D-glucose derivatives prepared in the same way, which showed retention times of 34.8 and 34.0 min, respectively.

ASSOCIATED CONTENT

Supporting Information

Full isolation procedures and 1D and 2D NMR spectroscopic data of **1**−**5** are available including ^{1}H, ^{13}C, COSY, HSQC, HMBC, and NOESY. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

ACKNOWLEDGMENTS

The authors thank Mr. S. Abbas (Ole Miss) for his assistance with plant collections, together with Kraft Foods and the National Institutes of Health (1R01AT007318) for some financial support. We also thank Dr. P. Griffin at the Scripps Research Institute Florida for performing the PPARγ agonistic activity evaluation, and Drs. A. Robbins and T. Schulz at Viacyte Inc. for cytotoxicity testing. This research was supported by National Institutes of Health grants (P41GM079597 and P01GM085354) and by the Scientific Research Program Funded by Shaanxi Provincial Education Department in China (2010JK749). This research was also supported in part by Xi'an University of Technology Starting

grant (108-211409), awarded to Dr. Y. He, and the Youth Scientists Innovation Team Program.

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