Three New Phenol Compounds from *Iris dichotoma* PALL.

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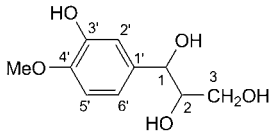
Three new phenolic compounds, irisdichototins D, E, and F (**1**–**3**, resp.) were isolated from the stems of *Iris dichotoma*. The structures of the new compounds were elucidated by spectroscopic analyses, including 2D-NMR techniques.

Introduction. – *Iris dichotoma* PALL. (Iridaceae), a perennial herb, used as folk-medicinal herb, is mainly distributed in the north of China. Its dried rhizomers, called '*Bai-Shegan*' in parts of China, are commonly used in Chinese folk medicine for clearing heat and detoxifying, and to eliminate phlegm, swelling, and pain in the throat [1]. Iridichototins A–C had been previously reported from the rhizomes of *I. dichotoma* [2]. To find potential drug leads, the chemical constituents of *I. dichotoma* were further investigated. Here, the isolation and structure elucidation of three new phenolic compounds is described.

Results and Discussion. – Repeated column chromatography of the EtOH extract from the rhizomes of *I. dichotoma* yielded irisdichototins D–F (**1**–**3**, resp.).



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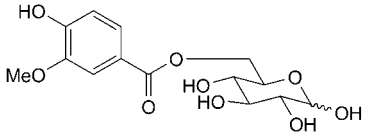

2 α-OH
3 β-OH

Irisdichototin D (**1**) was obtained as a white powder and assigned the molecular formula $C_{10}H_{14}O_5$ on the basis of HR-ESI-MS (*m/z* 237.0735 ($[M+Na]^+$)). The ^{1}H-NMR spectrum (*Table 1*) showed signals corresponding to two O-bearing CH groups at δ(H) 4.39 (*d*, *J* = 6.0, H–C(1)) and 3.45–3.42 (*m*, H–C(2)), two H-atoms of HO–CH_2 at δ(H) 3.31 (*dd*, *J* = 6.0, 4.8, CH_2(3)), three *ABX* aromatic H-atoms at δ(H) 6.88 (*d*, *J* = 1.2, H–C(2′)), 6.69 (*d*, *J* = 8.4, H–C(5′)), 6.68 (*dd*, *J* = 1.2, 8.4, H–C(6′)), and one MeO group at δ(C) 3.73 (*s*). The ^{13}C-NMR data of **1** (*Table 1*), combined with HSQC spectrum, exhibited ten C-atom signals comprising three signals of C-atoms of a propanoid. Analyses of the ^{1}H- and ^{13}C-NMR data indicated that the structure of compound **1** was similar to that of 4-*O*-methylguaiacylglycerol [3]. The main

Table 1. *^{1}H- and ^{13}C-NMR Data of* **1** (at 600 and 150 MHz, resp., in CD_3OD, δ in ppm, J in Hz)

	δ(H)	δ(C)
H–C(1)	4.39 (*d*, *J* = 6.0)	72.83
H–C(2)	3.45 – 3.42 (*m*)	75.98
CH_2(3)	3.31 (*dd*, *J* = 6.0, 4.8)	62.62
C(1′)		134.33
H–C(2′)	6.88 (*d*, *J* = 1.2)	110.97
C(3′)		145.31
C(4′)		147.00
H–C(5′)	6.69 (*d*, *J* = 8.4)	114.74
H–C(6′)	6.68 (*dd*, *J* = 1.2, 8.4)	119.05
HO–C(3′)	8.88 (br. *s*)	
*Me*O–C(4′)	3.73 (*s*)	55.53

difference between the two compounds is that compound **1** contains a OH instead of an MeO group at C(3′). The long-range HMBCs (*Fig. 1*) between the aromatic H-atoms (δ(H) 6.88 and 6.68) and C(4′) confirmed the position of the MeO group. Comparison of the aforementioned data with those reported in [3] revealed that **1** is 1-(3-hydroxy-4-methoxyphenyl)propane-1,2,3-triol, which is a new compound.

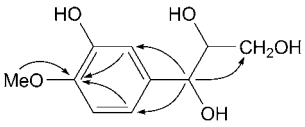

Fig. 1. *Key HMBCs of* **1**

Compound **1** was assumed to be *threo*-configured on the basis of the *J* value of 6.0 Hz of the benzylic H-atom (H–C(1)) signal at δ(H) 4.39 [4] and the CD spectrum with $Mo_2(OAc)_4$ (positive effects at 302 nm) [5]. However, the absolute configuration of **1** remains undetermined.

Irisdichototins E and F (**2** and **3**, resp.), white powder, were obtained as an epimeric mixture (*ca.* 1:1 by ^{1}H- and ^{13}C-NMR). The molecular formula $C_{14}H_{18}O_9$ was deduced from HR-ESI-MS (*m*/*z* 353.0918 ($[M + Na]^+$)). The ^{1}H-NMR spectrum (*Table 2*) of compound **2** exhibited signals corresponding to three *ABM* aromatic H-atoms at δ(H) 7.68 (*dd*, *J* = 1.2, 8.4), 7.55 (*d*, *J* = 1.2), and 6.84 (*dd*, *J* = 8.4), and a MeO group at δ(H) 3.87 (*s*), which were very similar to those of vanillic acid [6]. On the other hand, twelve H-atom signals were detected in the range of δ(H) 3.0 – 4.0, together with the signal at δ(H) 5.28 (*d*, *J* = 4.2) assigned to the anomeric H-atom, which suggested the presence of a sugar moiety. In combination with the ^{13}C-NMR spectrum, the signals of the C-atoms of the glucosyl moiety (C(1) to C(6)) were observed at δ(C) 92.10, 75.82, 74.18, 73.85, 69.70, and 63.47. In the HMBC spectrum of the compound (*Fig. 2*), the ester CO C-atom signal at δ(C) 168.7 was correlated with that of CH_2(6) of the glucose at δ(H) 4.54 (*dd*, *J* = 4.8, 12.6), indicating an ester linkage between the COO group and O–CH_2(6) moiety of the glucose. Acid hydrolysis of **2** gave glucose, which was identified by comparison with authentic sample. Based on these observations, the structure of **2** was established as shown.

Table 2. *^{1}H- and ^{13}C-NMR Data of* **2** *and* **3** (at 600 and 150 MHz, resp., in D_2O, δ in ppm, J in Hz)

	2		**3**	
	δ(H)	δ(C)	δ(H)	δ(C)
H–C(1)	5.28 (*d*, *J* = 4.2)	92.10	4.74 (*d*, *J* = 7.8)	96.08
H–C(2)	3.25 – 3.47 (*m*)	74.18	3.25 – 3.47 (*m*)	73.74
H–C(3)	3.25 – 3.47 (*m*)	73.85	3.25 – 3.47 (*m*)	75.61
H–C(4)	3.25 – 3.47 (*m*)	69.70	3.25 – 3.47 (*m*)	69.86
H–C(5)	3.25 – 3.47 (*m*)	75.82	3.25 – 3.47 (*m*)	77.54
CH_2(6)	4.50 – 4.54 (*dd*, *J* = 4.8, 12.6)	63.47	4.46 – 4.50 (*dd*, *J* = 5.4, 12.0)	63.47
C(1′)		168.65		168.65
C(1″)		116.82		116.82
H–C(2″)	7.55 (*d*, *J* = 1.2)	113.00	7.55 (*d*, *J* = 1.2)	113.00
C(3″)		148.91		148.91
C(4″)		156.37		156.37
H–C(5″)	6.84 (*d*, *J* = 8.4)	116.63	6.84 (*d*, *J* = 8.4)	116.63
H–C(6″)	7.68 (*dd*, *J* = 1.2, 8.4)	125.34	7.68 (*dd*, *J* = 1.2, 8.4)	125.34
*Me*O–C(3″)	3.87 (*s*)	55.90	3.87 (*s*)	55.90
HO–C(4″)	8.49 (*s*)		8.49 (*s*)	

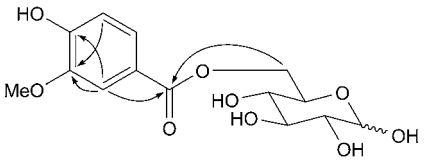

Fig. 2. *Key HMBCs of* **2** *and* **3**

Comparison of the ^{1}H- and ^{13}C-NMR data of **3** with those of **2** indicated that the small difference was that the β-glucose instead of α-glucose was attached to C(1′). The ^{1}H-NMR of **3** showed the signal at δ(H) 4.74 (*d*, *J* = 7.8) together with those of six H-atoms in the range of δ(H) 3.0 – 4.0. In the HMBC spectrum of compound **3** (*Fig. 2*), the CO C-atom signal at δ(C) 168.7 correlated that of CH_2(6) of the glucose at δ(H) 4.50 (*dd*, *J* = 5.4, 12.0), indicating the formation of an ester linkage between the COO group and the O–CH_2(6) moiety of the glucose. Therefore, the structure of **3** was established as shown.

Experimental Part

General. Prep. HPLC: *LabAlliance Series 1500* pump and a *LabAlliance Model 500* variable-wavelength detector with a reversed-phase (RP) column (*Alltech Alltima C18*, 5 μm, 10 × 250 mm, at 3.5 ml/min; detection at 260 nm). CD Spectra: *JASCO J-815* spectrophotometer. ^{1}H- and ^{13}C-NMR spectra (**1** in (D_6)DMSO and **2** in D_2O): *Bruker Avance-600* instrument, at 600 and 150 MHz, resp., chemical shifts δ in ppm with TMS as internal standard, coupling constants *J* in Hz. Mass spectra: *Thermo LTQ ORBITRAP XL* mass spectrometer (for ESI and HR-ESI).

Plant Material. The rhizomes of *I. dichotoma* were collected at Jiufeng mountain in Beijing, P. R. China, in April 2009, and identified by *P. X.*, Institute of Medicinal Plant Development, Chinese Academy of Medical Sciences and Peking Union Medical College. A voucher specimen (Bj090412) was deposited with the Institute of Medicinal Plant Development, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, P. R. China.

Extraction and Isolation. The air-dried rhizomes of *I. dichotoma* (5 kg) were refluxed with 90% EtOH (3 × 40 l, 2 h each). The extracts were concentrated under reduced pressure to syrup. The syrup was suspended in H_2O (6 l), followed by successive partitioning with petroleum ether (PE), AcOEt, and BuOH. The BuOH extract (326.3 g) was subjected to column chromatography (CC; SiO_2; $CHCl_3$/MeOH/H_2O 100 : 1 : 1 to 10 : 5 : 1) to afford twelve fractions, *Frs. 1–12*. *Fr. 1* (18.2 g) was further subjected to repeated CC ($CHCl_3$/MeOH/H_2O 99 : 1 : 1 to 10 : 3 : 1), and then with prep. HPLC (*Alltima C18*, 5 μm, 10 mm × 250 mm; MeOH/H_2O 43 : 57; flow rate 3.6 ml/min) to give compound **1** (9 mg) and a mixture of **2**/**3** (11 mg).

Acid Hydrolysis. Compound **2** or **3** (1 mg) was heated at 105° for 1 h in 1 ml of 2M HCl (H_2O/dioxane 1 : 1) separately and heated in a water bath at 80° for 2 h, then dioxane was evaporated, and the aglycone was removed by extracting with AcOEt (5 × 2 ml). The aq. layer was neutralized with Ag_2CO_3, centrifuged, and evaporated to dryness. The monosaccharide was identified as Glc by TLC (SiO_2; AcOEt/MeOH/H_2O/AcOH 13 : 2 : 1 : 3) comparison with authentic sugars.

Irisdichototin D (= rel-*(1*R,*2*S*)-1-(3-Hydroxy-4-methoxyphenyl)propane-1,2,3-triol*; **1**). White powder. $[\alpha]_D^{25} = +9.02$ ($c = 0.05$, MeOH). UV (MeOH): 228. CD ($c = 0.01$, DMSO + $Mo_2(OAc)_4$): 302 (+). IR (KBr): 3420, 1621, 1509, 1450. ^{1}H- and ^{13}C-NMR: see *Table 1*. HR-ESI-MS: 237.0735 ($[M + Na]^+$, $C_{10}H_{14}NaO_5^+$; calc. 237.0738).

Irisdichototins E and F (=*6*-O-*(4-Hydroxy-3-methoxybenzoyl)-α*-D-*glucopyranose and 6*-O-*(4-Hydroxy-3-methoxybenzoyl)-β*-D-*glucopyranose*; **2** and **3**, resp.). White powder. UV (MeOH): 275. IR (KBr): 3410, 1700, 1612, 1520, 1450. ^{1}H- and ^{13}C-NMR: see *Table 2*. HR-ESI-MS: 353.0918 ($[M + Na]^+$, $C_{14}H_{18}NaO_9^+$; calc. 353.0920).

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