Two New Chlorinated Naphthalene Glycosides from *Rumex patientia*

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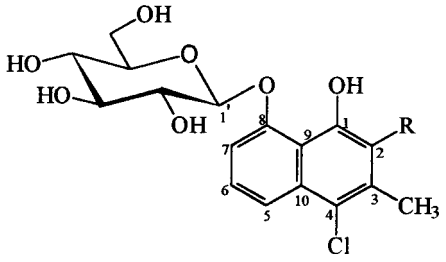
Two new naphthalene derivatives, named patientosides A (**1**) and B (**2**), were isolated from the roots of *Rumex patientia*. The structures of the new compounds were established as 2-acetyl-4-chloro-1,8-dihydroxy-3-methylnaphthalene-8-*O*-*β*-D-glucopyranoside (**1**) and 2,4-dichloro-1,8-dihydroxy-3-methylnaphthalene-8-*O*-*β*-D-glucopyranoside (**2**) by means of spectroscopic methods.

The genus *Rumex* (Polygonaceae) is represented by 25 species in the flora of Turkey.[1,2] The roots of *Rumex patientia* L. are reported to be used as a purgative and tonic in traditional medicine, and its leaves are commonly used as a green vegetable called "Labada" and "Develik".[3,4] Nine *Rumex* species including *R. patientia* have been attributed to the Chinese herbal medicine "Yangti", which has been used as a hemostatic and antifungal agent.[5] The roots of *R. patientia* are known to contain anthraquinone, tannin, naphthalene, and naphthoquinone derivatives.[6−8] The present paper describes the isolation and structure elucidation of two new chloro naphthalene glycosides, patientosides A (**1**) and B (**2**).



	R
1	$COCH_3$
2	Cl

Compound **1** was obtained as a pale yellow powder. It was detected on TLC by UV fluorescence (366 nm) and spraying with vanillin−H_2SO_4 (1%) reagent followed by heating at 100 °C for 5−10 min to give a blue-purple coloration. Its IR spectrum showed absorption bands due to hydroxyl groups (3369 cm^{-1}), a carbonyl group (1698 cm^{-1}), an aromatic moiety (1623 cm^{-1}), and an ether (1049 cm^{-1}). In the UV spectrum of **1**, absorption maxima were exhibited at 234, 312, and 339 nm. The molecular formula of **1** was determined to be $C_{19}H_{21}O_8Cl$ on the basis of the HREIMS. The EIMS of the compound **1** showed a very weak molecular ion peak at *m*/*z* 412, with a prominent fragment ion at *m*/*z* 250 corresponding to the loss of a glucosyl unit, and a peak at *m*/*z* 235 $[\text{aglycon} - CH_3]^+$. The results of the ESIMS also indicated a molecular mass of 412 and exhibited fragment peaks at *m*/*z* 399 $[(M - Cl) + Na]^+$ and 273 $[C_{13}H_{11}O_3Cl\ (\text{aglycon}) + Na]^+$. The ^1H NMR spectrum of **1** (Table 1) in CD_3OD exhibited the presence of two methyl groups at *δ* 2.37 and 2.59 as singlets and three aromatic protons at *δ* 7.99 (1H, dd, $J = 7.5$, 1.5 Hz, H-5), 7.52 (1H, dd, $J = 7.5$, 7.5 Hz, H-6), and 7.45 (1H, dd, $J = 7.5$, 1.5 Hz, H-7) with an ABC coupling pattern. It also revealed proton signals due to a sugar moiety between *δ* 3.44−5.14 including an anomeric proton (*δ* 5.14, 1H, d, $J = 8$ Hz, H-1′). The sugar was assigned as *β*-glucopyranose according to the ^1H and ^{13}C NMR data (Table 1).[9] ^{13}C NMR and APT spectra of **1** showed 19 resonance signals including eight methine, eight quaternary, two methyl, and one methylene carbon peak. Among these, a carbonyl group at *δ* 207.3 and two phenolic quaternary carbons at *δ* 156.3 (C-8) and 150.8 (C-1) could be assigned. The characteristic signals belonging to the sugar moiety were the following: one anomeric peak (C-1′ at *δ* 104.5), four aliphatic methine peaks (C-2′, C-3′, C-4′, C-5′ at *δ* 74.9, 78.1, 71.3, 78.9, respectively), and one aliphatic methylene carbon peak (C-6′ at *δ* 62.5). The connectivities between the protons on the naphthalene ring (H-5, H-6, and H-7) were apperent from the COSY and HMQC spectra. The HMBC spectrum (Figure 1) provided the substitution pattern of the naphthalene core and led to the connectivity between the sugar moiety and the aglycon on the basis of the cross-peak between C-8 (*δ* 156.3) and H-1′ (*δ* 5.14). Thus, the structure of **1** was determined to be 2-acetyl-4-chloro-1,8-dihydroxy-

Table 1. ^{13}C and ^1H NMR Spectral Data for Compounds **1** (CD_3OD) and **2** (DMSO-d_6)[a]

	1		**2**	
position	δ_C	δ_H, *J* (Hz)	δ_C	δ_H, *J* (Hz)
1	150.8		153.5	
2	127.1		116.2	
3	132.1		133.2	
4	122.9		120.3	
5	120.8	7.99 dd (7.5, 1.5)	119.0	7.89 d (8.4)
6	129.6	7.52 dd (7.5, 7.5)	128.2	7.59 dd (8.4, 7.8)
7	112.8	7.45 dd (7.5, 1.5)	111.9	7.50 d (7.8)
8	156.3		148.1	
9	115.9		114.3	
10	134.5		130.7	
CH_3-3	17.7	2.37 s	18.7	2.60 s
$CO\mathit{CH}_3$-2	32.5	2.59 s		
$\mathit{C}OCH_3$-2	207.3			
1′	104.5	5.14 d (8)	103.0	5.12 d (7.5)
2′	74.9	3.57 m	73.3	3.41 m
3′	78.1	3.55 m	76.2	3.39 m
4′	71.3	3.44 m	69.8	3.23 m
5′	78.9	3.55 m	77.8	3.45 m
6′	62.5	3.74 dd (12, 2)	60.7	3.54 m
		3.96 dd (12, 5.5)		3.78 dd (11, 6)

[a] The assigments were based on 1D and 2D NMR experiments.

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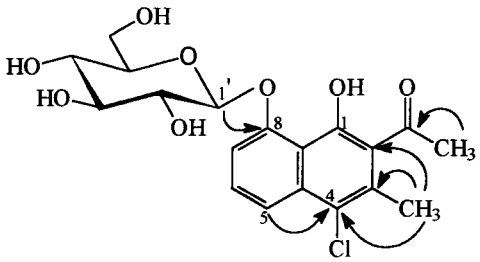

Figure 1. Selected heteronuclear multiple bond correlations of **1**.

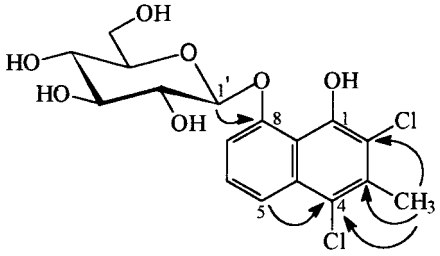

Figure 2. Selected heteronuclear multiple bond correlations of **2**.

3-methylnaphthalene-8-*O*-*β*-D-glucopyranoside (= 4-chloronepodin-8-*O*-*β*-D-glucopyranoside or 4-chloromusizin-8-*O*-*β*-D-glucopyranoside), to which the trivial name patientoside A has been attributed.

Compound **2** was obtained as a pale yellow powder and showed very a similar behavior by TLC to **1**. Its IR spectrum exhibited absorption bands due to hydroxyl groups (3379 cm^{-1}), an aromatic moiety (1609 cm^{-1}), and an ether (1034 cm^{-1}). In the UV spectrum of **2**, the absorption bands were observed at 235, 309, 325, and 341 nm. The molecular formula of the aglycon of **2** was determined to be $C_{11}H_8O_2Cl_2$ on the basis of HREIMS, and the DCIMS gave ions at *m*/*z* 422 $[M + NH_4]^+$, 180 $[C_6H_{10}O_5 + NH_4]^+$, and 197 $[180 + NH_3]^+$; the ion at *m*/*z* 422 was split into three peaks at *m*/*z* 422, 424, and 426 (relative intensities 1:5.8:9.1), revealing the isotopic pattern of the molecule due to the presence of two chlorine atoms.[10] The EIMS gave no molecular ion peak, but a fragment ion at *m*/*z* 242 $[M -$ glucose$]^+$ was observed. The ^1H NMR spectrum of **2** (Table 1) indicated the presence of a methyl group at δ 2.60 (3H, s), three aromatic protons at δ 7.89 (1H, d, J = 8.4 Hz, H-5), 7.59 (1H, dd, J = 8.4, 7.8 Hz, H-6), and 7.50 (1H, d, J = 7.8 Hz, H-7), an anomeric proton at δ 5.12 (1H, d, J = 7.5 Hz), and further signals typical for glucose. The anomeric configuration of the glucose was proposed to be β on the basis of the coupling constant (J = 7.5 Hz) and the chemical shift at δ 5.12.[9] The ^1H and ^{13}C NMR data of **2** (Table 1) were similar to those of **1** except for the presence of signals due to the acetyl moiety. In the COSY spectrum, aromatic protons represented the same spin system observed for **1**. The HMQC experiment permitted the assigments of the proton bearing carbon atoms. The glycosidic unit was determined to be attached to C-8, based on the long-range C−H coupling between C-8 (δ 148.1) and H-1′ (δ 5.12) in the HMBC spectrum (Figure 2). Therefore, compound **2** (patientoside B) was identified as 2,4-dichloro-1,8-dihydroxy-3-methylnaphthalene-8-*O*-*β*-D-glucopyranoside.

Chlorinated metabolites are often found in lichens, fungi, and marine algae but are not frequent in higher plants, where they mainly appear during the process of epoxide ring opening.[11] In most cases, these chlorine atoms are introduced during the workup or sometimes occur from natural sources. Chloro substituents on aromatic rings in plant metabolites are rare.[12,13] This is the first report of chlorinated metabolites from a plant in the genus *Rumex*.

Experimental Section

General Experimental Procedures. Optical rotations were recorded with Perkin-Elmer model 241 and 343 polarimeters using MeOH. UV spectra were determined in spectroscopic grade MeOH on a Shimadzu UV-160A spectrophotometer. IR spectra were measured on a Perkin-Elmer FTIR 1720 X spectrophotometer as pressed KBr disks. NMR spectra were recorded using a Bruker AMX 300 NMR spectrometer at 300 MHz for ^1H and 75.5 MHz for ^{13}C. Complete proton and carbon assignments were based on 1D (^1H, ^{13}C, and APT) and 2D ($^1H-^1H$ COSY, $^1H-^{13}C$ HMQC, and $^1H-^{13}C$ HMBC) NMR experiments. HREIMS, EIMS, ESIMS, and DCIMS were recorded on Varian MAT 731 and Finnigan MAT 311 A (EI 70 eV) instruments. TLC was performed on precoated silica gel 60 F_{254} aluminum sheets (0.2 mm, Merck). For column chromatography, normal-phase silica gel 60 (0.063−0.200 mm, Merck), reversed-phase silica gel (LiChroprep RP-18, Merck), and polyamide (Polyamid-MN-Polyamid SC 6, Macherey-Nagel, Düren) were used. Compounds were detected by UV fluorescence and/or spraying with vanillin−H_2SO_4 reagent followed by heating at 100 °C for 5−10 min.

Plant Material. The roots of *R. patientia* were collected from Bor, Niğde (1050 m in altitude), Turkey, in September 1996. The plant was identified by L.Ö.D. A voucher specimen has been deposited at the herbarium of the Faculty of Pharmacy, Hacettepe University, Ankara, Turkey (HUEF 96003).

Extraction and Isolation. Powdered dried plant material (500 g) was extracted with MeOH (3 × 2.5 L) at 40 °C. The MeOH extracts were combined and evaporated to dryness under a vacuum (100 g). The crude MeOH extract (40 g) was fractionated by open column chromatography on polyamide by gradient elution with H_2O−MeOH mixtures. The fractions eluted with 60% MeOH in water were rechromatographed over a polyamide column, eluting with H_2O−MeOH (60:40→20:80). The fractions eluted with 50% MeOH (1015 mg) were subjected to Si gel column chromatography, eluting with a $CHCl_3$−MeOH−H_2O gradient (90:10:0; 90:10:1; 80:20:2) to yield eight main fractions (fractions 1−8). Fraction 6 was applied to medium-presure liquid chromatography with LiChroprep RP-18, using a H_2O−MeOH gradient solvent system (40−70% MeOH). The fractions eluted with 60% MeOH and 70% MeOH were repeatedly chromatographed on Si gel eluting with $CHCl_3$−MeOH−H_2O (90:10:1) to yield compound **1** (36 mg) and compound **2** (25 mg), respectively.

Patientoside A (1): amorphous pale yellow powder; $[\alpha]^{20}_D$ −109.7° (*c* 0.75, MeOH); UV (MeOH) λ_{max} (log ϵ) 234 (3.97), 312 (3.73), 339 (3.69) nm; IR (KBr) ν_{max} 3369, 1698, 1623, 1049 cm^{-1}; ^1H NMR (CD_3OD, 300 MHz), see Table 1; ^{13}C NMR (CD_3OD, 75.5 MHz), see Table 1; EIMS *m*/*z* 412 $[M]^+$, 250 $[M -$ glucose$]^+$, 235 [aglycon − $CH_3]^+$; ESIMS *m*/*z* 412 $[M]^+$, 399 [(M − Cl) + Na]$^+$, 273 $[C_{13}H_{11}O_3Cl$ (aglycon) + Na$]^+$; HREIMS *m*/*z* 412.0924 (calcd for $C_{19}H_{21}O_8Cl$ 412.0924).

Patientoside B (2): amorphous pale yellow powder; $[\alpha]^{20}_D$ −235.8° (*c* 0.72, MeOH); UV (MeOH) λ_{max} (log ϵ) 235 (3.96), 309 (3.77), 325 (3.69), 341 (3.62) nm; IR (KBr) ν_{max} 3379, 1609, 1034 cm^{-1}; ^1H NMR (CD_3OD, 300 MHz), see Table 1; ^{13}C NMR (CD_3OD, 75.5 MHz), see Table 1; DCIMS *m*/*z* 422 $[M + NH_4]^+$, 180 $[C_6H_{10}O_5 + NH_4]^+$, 197 $[180 + NH_3]^+$, HREIMS *m*/*z* 241.9901 (calcd for $C_{11}H_8O_2Cl_2$ (aglycon), 241.9901).

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