Chemistry of Natural Compounds, Vol. 35, No. 1, 1999

TRITERPENE GLYCOSIDES OF *Hedera canariensis* IV. STRUCTURES OF GLYCOSIDES L-F_3, L-G_0, AND L-G_{1a} FROM THE LEAVES OF ALGERIAN IVY

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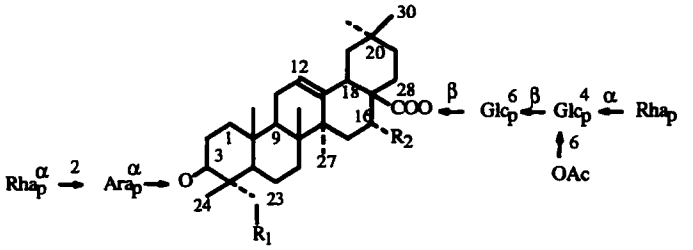
UDC 547.918:543.422

Three minor partially acetylated glycosides have been isolated from the leaves of Algerian ivy, Hedera canariensis Willd. (Araliaceae) — the previously known [3-O-[α-L-rhamnopyranosyl-(1→2)-O-α-L-arabinopyranoside] 28-O-[α-L-rhamnopyranosyl-(1→4)-O-(6-acetyl-β-D-glucopyranosyl)-(1→6)-O-β-D-glucopyranoside]s of oleanolic acid and of hederagenin (ciwujianoside C_4 and kizuta saponin K_{11}) and the new 3-O-[α-L-rhamnopyranosyl-(1→2)-α-O-L-arabinopyranoside] 28-O-[α-L-rhamnopyranosyl-(1→4)-O-(6-O-acetyl-β-D-glucopyranosyl)-(1→6)-O-β-D-glucopyranoside of echinocystic acid (glycoside L-G_0). The structures of the glycosides isolated have been established on the basis of chemical transformations and ^1H and ^{13}C NMR spectroscopy.

In the present paper we describe the isolation of three minor partially acetylated glycosides from the leave of Algerian ivy, *Hedera canariensis* Willd. (Araliaceae) and the determination of their structures. These glycosides were detected in fractions L-F and L-G [1]. We have described the isolation of glycoside L-F_3 (**1**) previously [2], and on the chromatographic separation of fraction L-G we obtained the least polar individual glycoside predominating in it, L-G_0 (**2**), while in the chromatographic purification of glycoside L-G_1 we isolated the accompanying glycoside L-G_{1a} (**3**).



1. R_1=H, R_2=H
2. R_1=H, R_2=OH
3. R_1=OH, R_2=H

According to the results of complete acid hydrolysis, the carbohydrate compositions of (**1**—**3**) were represented by identical sets of sugars — rhamnose, arabinose, and glucose —, while the aglycons were oleanolic and echinocystic acids and hederagenin, respectively. Cleavage of the acylglycosidic bonds in (**1**—**3**) under the conditions of severe alkaline hydrolysis gave progenins identical, according to TLC in various solvent systems, with the glycosides L-C, L-D, and L-E_1 previously isolated from Algerian ivy leaves [1], which are the 3-O-[α-L-rhamnopyranosyl-(1→2)-O-α-L-arabinopyranoside]s of oleanolic and echinocystic acids and hederagenin.

Thus, (**1**—**3**) are bisdesmosides and the carbohydrate chains at the C-3 atoms of the aglycons are represented by the same disaccharide fragment, α-*L*-Rha*p*-(1→2)-α-*L*-Ara*p*-, which is typical for the glycosides of plants of the ivy (*Hedera*) genus.

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0009-3130/99/3501-0065$22.00

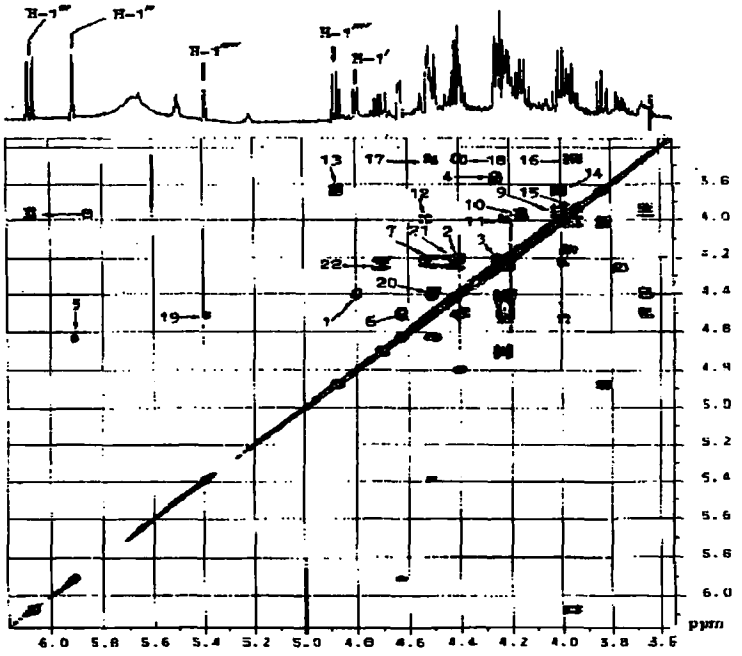

Fig. 1. COSY two-dimensional spectrum of glycoside L-G_0 (400 MHz). The figures show cross-peaks between the following protons: *1* - (1, 2)′; *2* - (2, 3)′; *3* - (3, 4; 4, 5e)′; *4* - (4, 5a; 5a, 5e)′; *5* - (1, 2)″; *6* - (2, 3)″; *7* - (3, 4; 4, 5)″; *8* - (1, 2)‴; *9* - (2, 3)‴; *10* - (3, 4; 4, 5)‴; *11* - (5, 6B)‴; *12* - (5, 6A)‴; *13* - (1, 2)⁗; *14* - (2, 3)⁗; *15* - (3, 4)⁗; *16* - (4, 5)⁗; *17* - (5, 6A)⁗; *18* - (5, 6B)⁗; *19* - (1, 2)⁗′; *20* - (2, 3)⁗′; *21* - (3, 4)⁗′; *22* - (4, 5)⁗′.

The mild alkaline hydrolysis of (**1**—**3**) with an aqueous alcoholic solution of ammonia enabled us to obtain from them the more polar glycosides L-G_2, L-H_1, and L-H_2, isolated from the leaves of this plant previously [1] and consisting of the {3-O-[α-L-rhamnopyranosyl-(1→2)-O-α-L-arabinopyranoside] 28-O-[α-L-rhamnopyranosyl-(1→4)-O-β-D-glucopyranosyl-(1→6)-O-β-D-glucopyranoside}s of oleanolic and echinocystic acids and of hederagenin. This determined partial structures of (**1**—**3**) and presupposed the presence of additional acyl groups in them.

The question of the number, nature, and localization of the acyl groups was solved by the use of ^{1}H and ^{13}C NMR spectra. In the PMR spectra of (**1**—**3**), in addition to the signals of the protons of the carbohydrate residues, the assignment of which was made with the help of COSY two-dimensional spectroscopy (Fig. 1); in each case one three-proton singlet signal was observed at δ 1.90 ppm, in the usual region for acetates (1.80—2.20 ppm). In each of the ^{13}C NMR spectra of (**1**—**3**), as compared with the spectra of the above-mentioned glycosides L-G_2, L-H_1, and L-H_2, we found two additional signals of C atoms, with δ 21.2 and 171.6 ppm, which also confirmed the presence of acetyl groups in glycosides (**1**—**3**). The positions of the acetyl groups in (**1**—**3**) were established by comparing the NMR subspectra of the carbohydrate moieties of (**1**—**3**) with the subspectra of the nonacetylated fragments α-*L*-Rha*p*-(1→2)-α-*L*-Ara*p*→ and →β-*D*-Glc*p*-(6→1)-β-*D*-Glc*p*-(4→1)-α-*L*-Rha*p* [2—6]. At the same time, in the PMR spectra of (**1**—**3**) we observed shifts of the signals of H-5″″ (+0.1 ppm), H-6A″″ (+0.4 ppm), and H-6B″″ (+0.4 ppm), and in the ^{13}C NMR spectra those of C-6″″ (+3.7 ppm) and C-5″″ (−3.3 ppm),together with a small effect on C-4″″ (+1.0 ppm), which unambiguously determined the position of the O-acetyl group at the C-6″″ atom of the Glc″″ residue in each case. In addition, the chemical shifts of the C-atoms of the trisaccharide chain coincided completely with those for the fragment →β-*D*-Glc*p*-(6→1)-(6-OAc-β-*D*-Glc*p*)-(4→1)-α-*L*-Rha*p* [4—6].

The types of bonds in the carbohydrate fragments of (**2**) were additionally confirmed by an analysis of the PMR spectrum of the full acetate (**2a**), where in 3.5—4.0 ppm region besides usually presented signals of H-5 monosacchride residues we observed the signals of H-2 of Ara′, H-6A and -6B of Glc‴, and H-4 of Glc″″. The assignment of the signals in the PMR spectrum of (**2a**) was made on the basis of a COSY two-dimensional spectrum (Fig. 2).

TABLE 1. Chemical Shifts of the Signals of the ^{13}C Atoms of the Aglycon Moiety of Glycoside L-G_0 (**2**) (δ, ppm, 0 — TMS, C_5D_5N)

C atom	Chem. shift	C atom	Chem. shift	C atom	Chem. shift	C atom	Chem. shift
1	39.5	9	47.7	17	49.7	25	16.2
2	27.0	10	37.5	18	41.8	26	18.0
3	89.4	11	24.3	19	47.7	27	27.7
4	40.0	12	123.1	20	31.2	28	176.7
5	56.5	13	145.0	21	36.4	29	33.6
6	18.6	14	42.5	22	32.4	30	25.2
7	33.8	15	36.4	23	28.6		
8	40.5	16	74.1	24	17.4		

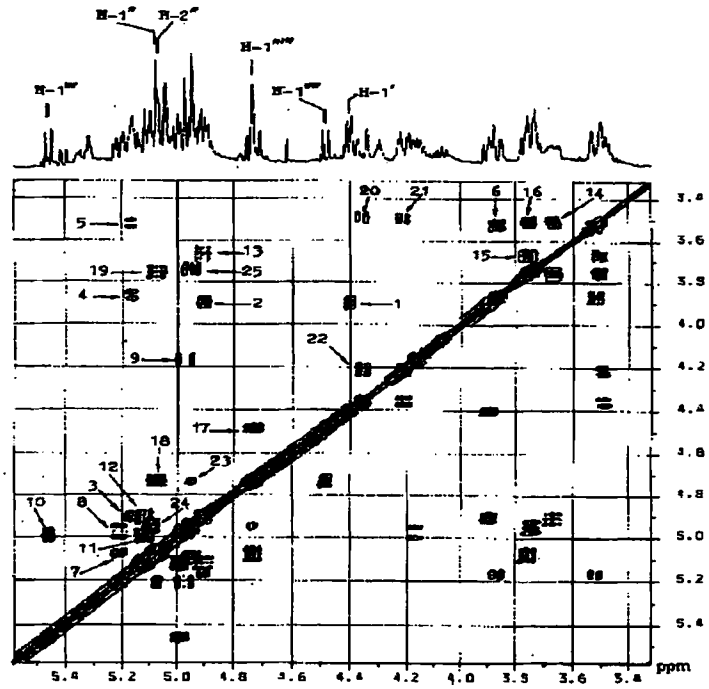

Fig. 2. COSY two-dimensional spectrum of the full acetate of glycoside L-G_0 (400 MHz). The figures show cross-peaks between the following protons: *1* - (1, 2)′; *2* - (2, 3)′; *3* - (3, 4)′; *4* - (4, 5e)′; *5* - (4, 5a)′; *6* - (5a, 5e)′; *7* - (2, 3)″; *8* - (3, 4)″; *9* - (4, 5)″; *10* - (1, 2)‴; *11* - (2, 3)‴; *12* - (3, 4)‴; *13* - (4, 5)‴; *14* - (5, 6B)‴; *15* - (5, 6A)⁗; *16* - (6A, 6B‴; 4, 5 ‴); *17* - (1, 2)⁗; *18* - (2, 3)⁗; *19* - (3, 4)⁗; *20* - (5, 6A)⁗; *21* - (5, 6B)⁗; *22* - (6A, 6B)⁗; *23* - (1, 2)⁗′; *24* - (2, 3; 3, 4)⁗′; *25* - (4, 5)⁗′.

Thus, the glycosides isolated, L-F_3, L-G_0, and L-G_{1a}, were the 3-O-[α-L-rhamnopyranosyl-(1→2)-O-α-L-arabinopyranosyls of the 28-O-[α-L-rhamnopyranosyl-(1→4)-O-(6-O-acetyl-β-D-glucopyranosyl)-(1→6)-O-β-D-glucopyranosyl esters of oleanolic acid, of echinocytic acid, and of hederagenin, respectively. Glycoside L-G_0 is a new triterpene glycoside, glycoside L-F_3 is identical in structure with civuyanoside C_4 from the leaves of *Acanthopanax senticosus* [5], and glycoside L-G_{1a} with kizutasaponin K_{11} from the leaves of *Kalopanax pictus* [4] and *Hedera rhombea* [6].

TABLE 2. Chemical Shifts of the Signals of the ^{13}C Atom of the Carbohydrate Moiety of Glycoside L-G_0 (2) (δ, ppm, 0 — TMS, C_5D_5N)

C atom	Chem. shift	C atom	Chem. shift
Ara ′		Glc ′′′	
1	105.0	1	96.1
2	76.5	2	74.0
3	74.1	3	78.6
4	68.6	4	71.0
5	64.5	5	78.3
		6	69.6
Rha ′′		Glc ′′′	
1	102.1	1	104.9
2	72.5	2	75.3
3	72.7	3	76.5
4	74.5	4	79.6
5	70.4	5	73.9
6	18.8	6	64.1
		Rha ′′′	
		1	103.1
		2	72.5
		3	72.7
		4	74.0
		5	70.9
		6	18.9

EXPERIMENTAL

For general observations and the isolation of fractions L-F and L-G, see [1, 2].

NMR spectra were obtained with Bruker WM-250 and AM-400 instruments.

Glycosides solutions were used in C_5D_5N and $CDCl_3$.

The chromatographic separation of 400 mg of fraction L-G on silica gel (SiO_2) with elution by the water-saturated chloroform—ethanol (2:1) system gave 55 mg of the pure glycoside (2) and glycosides L-G_1—L-G_4 [1]. TLC analysis of glycoside L-G_1 showed that it contained a minor glycoside with a lower chromatographic mobility, which we have designated as L-G_{1a} (3). Glycoside (3) was obtained in the individual state (15 mg) on the additional purification of L-G_1 on Silpearl microspherical silica gel with elution by water-saturated chloroform—ethanol (2:1).

Compound (2) was acetylated with acetic anhydride in pyridine (1:1) at 20°C for 12 h, followed by evaporation after the addition of benzene.

Glycoside L-F_3 (1), $[\alpha]_D$ -20°(*c* 0.5; methanol; lit. $[\alpha]_D$ -23.7°(methanol) [5]. In a complete acid hydrolysate of (1) by TLC we identified rhamnose, arabinose, glucose, and oleanolic acid. The alkaline hydrolysis of (1) gave glycoside L-C [1], while its mild alkaline hydrolysis led to glycoside L-G_2 from Algerian ivy leaves [1].

The ^{13}C NMR spectrum of glycoside (1) was identical with that of ciwujianoside C_4 [5]. The ^1H subspectra of the carbohydrate fragments of (1) were identical with those for (2).

Glycoside L-G_0 (2), $[\alpha]_D$ -42° (*c* 1.0, pyridine). In a full acid hydrolysate of (2) we found rhamnose, arabinose, glucose, and echinocystic acid. According to TLC, a progenin from (2) was identical with glycoside L-D from Algerian ivy leaves [1]. The mild acid hydrolysis of (2) gave the glycoside L-H_1 [1].

PMR spectrum of (2) (δ, ppm, 0-TMS, C_5D_5N): 4.80 (d, H-1′, $J_{1,2}$=5.5 Hz), 4.40 (t, H-2′, $J_{2,3}$=6.5 Hz), 4.20-4.27 (m, H-3′, H-4′, H-5e′), 3.75 (m, H-5a′), 5.91 (d, H-1′′, $J_{1,2}$=1.5 Hz), 4.63 (d, H-2′′, $J_{2,3}$=3.5 Hz), 4.50 (dd, H-3′′, $J_{3,4}$=9.5 Hz), 4.23 (t, H-4′′, $J_{4,5}$=9.5 Hz), 4.50 (m, H-5′′), 1.58 (d, H-6′′, $J_{5,6}$=6.5 Hz), 6.08 (d, H-1′′′, $J_{1,2}$=8.0 Hz), 3.96 (t, H-2′′′, $J_{2,3}$=8.5 Hz),

4.01 (t, H-3‴, $J_{3,4}$=9.0 Hz), 4.17 (t, H-4‴, $J_{4,5}$=9.0 Hz); 4.00 (m, H-5‴), 4.53 (H-6A‴), 4.23 (H-6B‴), 4.87 (d, H-1⁗, $J_{1,2}$=8.0 Hz), 3.83 (t, H-2⁗, $J_{2,3}$=8.5 Hz), 3.92-4.04 (m, H-3⁗, H-4⁗), 3.66 (m, H-5⁗), 4.50 (H-6A⁗), 4.39 (H-6B⁗), 5.39 (d, H-1⁗⁗, $J_{1,2}$=1.5 Hz), 4.51 (dd, H-2⁗⁗, $J_{2,3}$=3.5 Hz), 4.41 (dd, H-3⁗⁗, $J_{3,4}$=9.0 Hz), 4.23 (t, H-4⁗⁗; $J_{4,5}$=9.5 Hz), 4.70 (dq, H-5⁗⁗, $J_{5,6}$=6.5 Hz), 1.52 (d, H-6⁗⁗), 3.28 (dd, H-3, $J_{2e,3}$=3.5 Hz, $J_{2a,3}$=12.0 Hz), 5.22 (t, H-12, $J_{11,12}$=3.5 Hz), 5.50 (t, H-16, $J_{15,16}$=3.5 Hz), 3.14 (dd, H-8, $J_{18,\ 19e}$=4.0 Hz, $J_{18,\ 19a}$=13.5 Hz), 1.70, 1.05, 1.00, 0.97, 0.94, 0.88, 0.80 (all s, 7CH_3), 1.90 (s, -$COCH_3$).

The ^{13}C NMR spectrum of (2) is given in Tables 1 and 2.

Full Acetate of Glycoside L-G_0 (2a), $[\alpha]_D$ -11° (*c* 0.5, chloroform), lit. $[\alpha]_D$ -13.2° (*c* 2.1; chloroform) [3].

PMR spectrum of (2a) (δ, ppm, 0-TMS, $CDCl_3$): 4.40 (d, H-1′, $J_{1,2}$=6.5 Hz), 3.90 (dd, H-2′, $J_{2,3}$=8.5 Hz), 4.90(dd, H-3′, $J_{3,4}$=4.0 Hz), 5.16 (m, H-4′), 3.52 (dd, H-5a′, $J_{4,5a}$=2.5 Hz), 3.87 (dd, H-5e′, $J_{5a,5e}$=14.0 Hz, $J_{4,5e}$=4.0 Hz), 5.07 (d, H-1″, $J_{1,2}$=1.5 Hz), 5.07 (dd, H-2″, $J_{2,3}$=3.5 Hz), 5.21 (dd, H-3″, $J_{3,4}$=10.0 Hz), 4.97 (t, H-4″, $J_{4,5}$=10.0 Hz), 4.20 (dq, H-5″), 1.12 (d, H-6″, $J_{5,6}$=6.5 Hz), 5.46 (d, H-1‴, $J_{1,2}$=8.5 Hz), 4.99 (t, H-2‴, $J_{2,3}$=8.5 Hz), 5.12 (t, H-3‴, $J_{3,4}$=9.5 Hz); 4.91 (t, H-4‴, $J_{4,5}$=10.0 Hz), 3.66 (m, H-5‴), 3.74 (dd, H-6A‴, $J_{6A,6B}$=12.0 Hz),3.49 (dd, H-6B‴), 4.48 (d, H-1⁗, $J_{1,2}$=8.0 Hz), 4.73 (dd, H-2⁗, $J_{2,3}$=9.5 Hz), 5.07 (t, H-3⁗, $J_{3,4}$=9.5 Hz), 3.76 (t, H-4⁗, $J_{4,5}$=9.5 Hz), 3.49 (m, H-5⁗), 4.36 (dd, H-6A⁗, $J_{5,6A}$=2.0 Hz, $J_{6A,6B}$=13.0 Hz), 4.21 (dd, H-6B⁗, $J_{5,6B}$=4.0 Hz), 4.73 (d, H-1⁗⁗, $J_{1,2}$=1.5 Hz), 4.95 (dd, H-2⁗⁗, $J_{2,3}$=4.0 Hz), 5.09 (dd, H-3⁗⁗, $J_{3,4}$=10.0 Hz), 4.95 (t, H-4⁗⁗, $J_{4,5}$=10.0 Hz), 3.74 (dq, H-5⁗⁗, $J_{5,6}$=6.5 Hz), 1.06 (d, H-6⁗⁗), 3.07 (dd, H-3, $J_{2e,3}$=4.0 Hz, $J_{2a,3}$=12.0 Hz), 4.30 (br.t, H-12, $J_{11,12}$=3.5 Hz), 5.32 (br.t, H-16, $J_{15,16}$=3.5 Hz), 2.87 (dd, H-18), 1.26, 0.96, 0.95, 0.86, 0.82, 0.76, 0.67 (all s, 7CH_3).

Glycoside L-G_{1a} (3), $[\alpha]_D$ -25° (*c* 0.5, methanol), lit. $[\alpha]_D$ -22.1° (methanol) [4]. In a complete acid hydrolysate of (3) we identified rhamnose, arabinose, glucose, and hederagenin. A progenin from (3) obtained by alkaline hydrolysis was identical, according to TLC, with glycoside L-E_1 [1] while the mild alkaline hydrolysis of (3) gave glycoside L-H_2 [1].

The ^{13}C NMR spectrum of glycoside (3) was identical with that of kizuta saponin K_{11} [4, 6]. The chemical shifts of the protons of the disaccharide fragment of (3) coincided with those of glycosides L-E_2 and L-H_3 from Algerian ivy leaves [7], and those of the trisaccharide fragment with those of (2).

The work was partially financed by The International Soros Science Education Program (ISSEP), grant No. APU073024 and grant No. PSU083096.

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