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New water soluble flavone and xanthone glycosides from *Hypericum canariense* L.

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ABSTRACT

The water soluble portion of the aerial parts of *Hypericum canariense* L. yielded after acetylation the 5,7,3′4′-tetra- and 7,3′4′-triacetates of a new flavonoid 5,7,3′,4′-tetrahydroxy-3-*O*-β-D-(methyl 2,3,4-triacetoxypyranuronyl)-quercetin, the 3′-acetate of a new flavonoid 3′-hydroxy-5,7,4′-trimethoxy-3-*O*-β-D-(methyl 2,3,4-triacetoxypyranuronyl)-quercetin, the 3′-acetate and the 3′5′-diacetate of the new flavonoid 5,3′dihydroxy-7,4′-dimethoxy-3-β-D-(methyl 2,3,4-triacetoxypyranuronyl)-quercetin, the xanthone derivative mangiferin 2′,3′,4′,6′-tetraacetate and the latter's new 1,3,6,7′-tetramethoxy, 1,3,6-trimethoxy-4-acetoxy and 1,7-diacetoxy-3,6-dimethoxy analogs.

1. Introduction

Chloroform extracts of *Hypericum canariense* L. (Hypericaceae), an endemic of the Canary Islands, have yielded a number of xanthones as well as the flavonoids quercetin, hyperoside and 3,8′-biapigenin (Cardona et al., 1985, 1986, 1989) while the flavonoids kaempferol, quercetin and hyperoside have apparently also been detected in this species (Makovetskaya, 2001). Recent articles report that methanol extracts of *Hypericum canariense* exhibit various types of biological activity (Prado et al., 2002; Rabanal et al., 2002, 2005; Sanchez-Mateo et al., 2002). Since aerial parts of *Hypericum canariense* have been used as decoctions in the Canary Islands (Perez de Paz and Hernandez Padron, 1999) further investigation of the polar constituents of this species seemed warranted and we therefore attempted to establish the identity of biologically active constituent(s) from the water soluble fraction which had not been investigate previously. Isolated after acetylation of the complex mixture to permit more facile separation of the polar components and extensive chromatography were the new polyacetylated flavonol methyl glucopyranuronates **1**–**5**, the known xanthone derivative **6** (mangiferin 2′,3′,4′,6′-tetraacetate, Faizi et al., 2006) and the latter's new 1,3,6-trimethoxy-7-acetoxy- (**8**) and 1,7-diacetoxy-3,6-dimethoxy analog **9**. We also encountered 1,3,6,7-tetramethoxy derivative **7** which has been prepared previously by partial synthesis (Aritomi and Kawasaki, 1970) although its characterization was sparse (Fig. 1).

2. Results and discussion

The molecular formula of compound **1**, MP 197–199 °C, was established as $C_{36}H_{34}O_{20}$ by means of its HRSI-TOF MS $[M + Na]^+$ peak at *m*/*z* 809.1545. The ^1H spectrum of **1** (Table 1, ^{13}C NMR spectrum in Table 2) contained aromatic, glycosidic, methoxyl and acetyl protons. Two *meta*-coupled resonances at δ_H 6.84 (1H, d, J = 2.2 Hz, δ_C 113.5), 7.29 (1H, d, J = 2.2 Hz, δ_C 108.9) were assigned to H-6 and H-8 of the flavonoid A ring. The remaining aromatic resonances at δ_H 8.06 (1H, d, J = 2.1 Hz, δ_C 124.9), 7.33 (1H, d, J = 8.5 Hz, δ_C 123.3) and 7.93 (1H, dd, J = 8.5, 2.2 Hz, δ_C 127.2) were assigned to H-2′, H-5′ and H-6′ respectively. Analysis of correlations in the HSQC and HMBC spectra provided the full assignment for the aglycone part, i.e. quercetin, of substance **1**. Long range correlations in the HMBC spectrum between the resonance of the anomeric proton at δ_H 5.68 (1H, d, J = 7.9 Hz, δ_C 98.6) and C-3 of the aglycone at δ_C 136.4 provided proof for 3-*O*-glycosylation. The large coupling constant of the anomeric proton was consistent with a β-glycosidic linkage. The sugar portion contained a five proton spin system in which H-5g appeared as a doublet at δ_H 3.97 (1H, d, J = 9.85 Hz, δ_C = 72.3) with only one vicinal correlation in the DQSFCOSY spectrum instead of the usual ddd pattern for H-5′. Additionally an HMBC correlation between H-5g and the carboxyl carbon at δ_C 166.7 indicated that C-6g was oxidized thus establishing that a glucopyranosiduronic acid unit was attached to C-3 in compound **1**. A further HMBC correlation between the three proton singlet at δ_H 3.61 (δ_C 52.7) and the former carboxylic carbon permitted placement on C-6 g of **1** of a methoxy group. Substance **1** was synthesized a number of years ago (Wagner et al., 1970) although ^{13}C NMR data were lacking.

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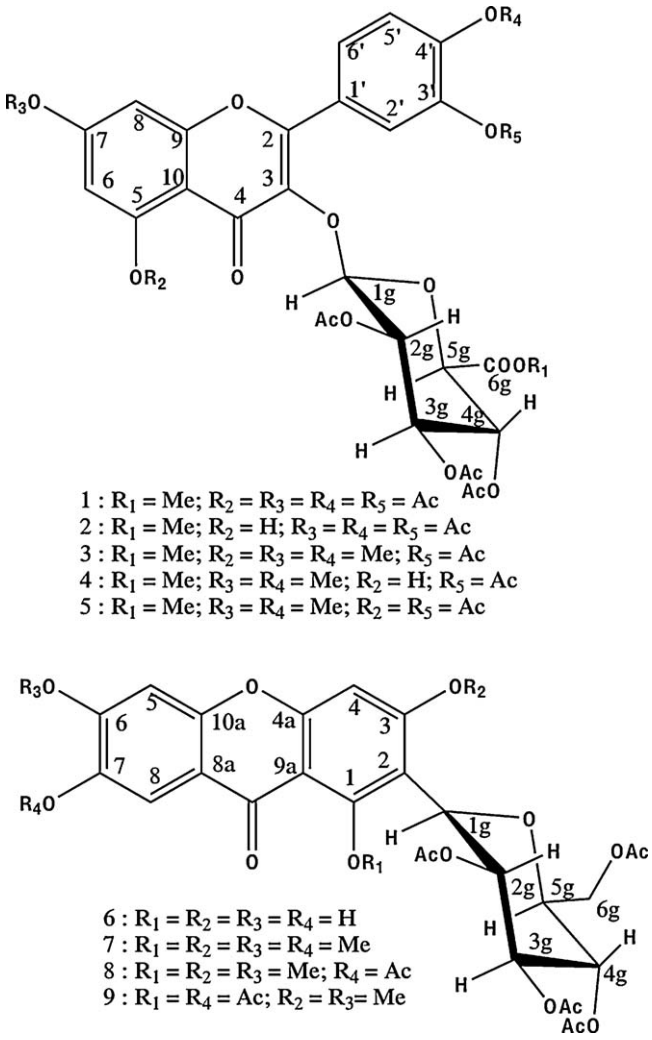

Fig. 1. Flavone and xanthone glycosides from *Hypericum canariense* L.

The ^{1}H and ^{13}C NMR spectra of the second flavonoid **2** (Tables 1 and 2) closely resembled those of **1** except for the presence in the NMR spectrum of **2** of the signal of a hydrogen bonded hydroxyl group at δ_H 12.33 which had replaced the signals of the acetate formerly on C-5, an observation corroborated by the HRMS. All assignments here and subsequently, were, as in the case of **1**, based on HSQC and HMBC correlations and will not be discussed in detail.

The remaining flavonoids **3**–**5** were only obtained after methylation of a mixture to improve separation, by chromatography, of a partially acetylated inseparable mixture. The HRMS of **3** corresponded to molecular formula $C_{33}H_{34}O_{17}$. Its ^{1}H and ^{13}C NMR spectra (Tables 1 and 2) resembled those of **1** but contained three additional –OMe groups. The C-5, C-7 and C-4′ hydroxyl groups of the quercetin parent were now methylated, with an acetate function on C-3′, whereas another substance **4** with a chelated hydroxyl obviously on C-5 and only two methoxyls on C-7 and C-4′· had a HRMS corresponding to $C_{32}H_{32}O_{17}$ as well as ^{1}H and ^{13}C NMR spectra (Tables 1 and 2) consonant with structure **4**. As for the remaining member **5** of this series of derivatives from *Hypericum canariense*, the HRMS corresponded to empirical formula $C_{34}H_{34}O_{18}$ while analysis of the ^{1}H and ^{13}C NMR spectra (Tables 1 and 2), which exhibited two acetate frequencies in addition to those of the pyranuronyl moiety in the manner detailed for **3** and **4** clearly showed that the substance was the 5,3′-diacetoxy-7,4′′-dimethoxy analog **5**. Locations of the acetoxy and methoxy groups were confirmed by HMBC and Roesy correlations.

The molecular formula of substance **6**, MP 138–140 °C, was established as $C_{27}H_{36}0_{15}$ by HRESIMS while the ^{1}H NMR spectrum (Table 3) indicated the presence of a 1,2,4,5-tetrasubstituted and one pentasubstituted aromatic ring as well as one tetraacetylated β-glucopyranosyl moiety. The ^{13}C NMR spectrum (Table 4) exhibited 27 signals including those of 12 aromatic carbons and one conjugated carbonyl. All of which suggested the presence of a xanthone. The chemical shift of a proton singlet at δ_H 8.43 was indicative of a proton *peri* to rhe carbonyl on a dioxygenated aomatic ring, an observation which implied that the proton of a chelated hydroxyl at δ_H 13.6 was part of the second ring of the xanthone nucleus. In the HMBC spectrum this signal also exhibited cross-peaks with three quaternary carbons at δ_C 160.5. 102.1 and l03.l, the latter correlating with the anomeric proton of the sugar moiety at δ_H 5.30 (J = 10) and with an aromatic proton at δ_H 6.32, all of which indicated that the chelated hydroxyl, the glucosyl unit, and the aromatic proton were located on the same ring of the xanthone unit. The chemical shift and coupling constant of the anomeric proton together with six resonances between δ 61.6 and δ 73.0 accorded with the attachment by a C–C bond of a β-glucopyranosyl unit to the aglycone while cross-peaks at δ_H 5.30/103.1 (H-1′/C-2), δ_H 5.30/160.5 (H-1′/C-1) and δ_H 5.30/

Table 1
^{1}H NMR spectroscopic data of flavonoid acetates **1**–**5** (600 MHz, $CDCl_3$).

Protón	1	2	3	4	5
H-6	6.84 d (2.2)	6.58d (2.2)	6.35 d (2.2)	6.36 d (2.1)	6.60 d (2.0)
H-8	7.29 d (2.2)	6.82 d (2.2)	6.49 d (2.2)	6.45 d (2.1)	6.81 d 2.0)
H′-2	8.06 d (2.1)	8.11 d (2.1)	7.82 d (2.1)	7.82 d (2.2)	7.79 d (2.0)
H′-5	7.33 d (8.6)	7.35 d (8.8)	7.07 d (8.8)	7.08 d (8.8)	7.07 d (8.8)
H′-6	7.93 dd (8.6, 2.1)	7.95 dd (8.8, 2.1)	8.0 dd (8.8, 2.1)	8.07 dd (8.8, 2.2)	8.04 dd (8.8, 2.0)
(OH) at C-5		12.33 **s**		12.41 s	
H-1g	5.68 d (7.9)	5.77 d (7.8)	5.90 d (7.8)	5.77 d (7.8)	5.68 d (7.7)
H-2g	5.22 dd (9.5, 7.9)	5.25 dd (9.6, 7.8)	5.23 dd (9.5, 7.8)	5.25 dd (9.5, 7.8)	5.21dd (9.7, 7.7)
H-3g	5.32 t (9.5)	5.37 t (9.5)	5.32 t (9.5)	5.37 t (9.5)	5.37 t (9.7)
H-4g	5.20 br t (9.6)	5.23br t (9.5)	5.20 br t (9.5)	5.23 br t (9.7)	5.19 br t (9.6)
H-5g	3.97 d (9.85)	4.0 d (10)	4.00 d (9.9)	3.99 d (9.9)	3.97 d (9.6)
OAc 2g	2.10	2.11	2.09	2.11	2.10
OAc 3g	2.04	2.04	2.02	2.04	2.04
OAc 4g	2.01	2.02	2.01	2.02	2.01
OAc (C-5)	2.46			–	2.45
OAc (C-7)	2.347	2.33		–	–
OAc (C′-3)	2.36	2.37	2.37	2.38	2.37
OAc (C′-4)	2.340	2.34			–
OMe (CO)	3.61	3.62	3.60 s	3.62	3.6
OMe (C-5)	–	–	3.98	–	–
OMe (C-7)	–	–	3.90	3.87	3.90
OMe (C′-4)	–	–	3.94	3.94	3.94

Table 2
^{13}C NMR spectra of flavonoid acetates **1**–**5**, $CDCl_3$, 150 MHz.

Carbon	1	2	3	4	5
C-2	154.7	156.6	153.7	156.4	154.7
C-3	136.4	134.7	135.6	133.8	135.7
C-4	171.8	178.0	172.9	177.4	171.7
C-5	150.1	161.7	160.9	161.8	150.3
C-6	113.5	105.3	96.1	98.2	108.4
C-7	153.9	156.2	164.3	165.6	163.4
C-8	108.9	101.1	92.5	92.2	98.5
C-9	156.5	155.7	158.8	156.7	157.7
C-10	115.0	109.1	109.3	105.8	111.2
C′-1	128.2	128.0	128.9	122.5	122.7
C′-2	124.9	125.2	123.3	123.5	123.3
C′-3	141.8	141.9	139.3	139.3	139.3
C′-4	144.0	144.4	152.9	153.4	153.1
C′-5	123.3	123.5	111.8	111.8	111.8
C′-6	127.2	127.3	128.5	128.8	128.6
C-1g	98.6	98.9	98.4	99.0	98.8
C-2g	71.2	71.2	71.3	71.3	71.3
C-3g	71.7	71.7	71.9	71.8	71.8
C-4	69.3	69.2	69.6	69.4	69.4
C-5g	72.3	72.5	72.4	72.4	72.3
C-6g	166.7	166.6	166.9	166.7	166.7
Ac (C-2g)	20.71,169.7	20.7, 169.6	20.8, 169.9	20.8, 169.7	20.8, 169.8
Ac (C-3g)	20.67,170.0	20.6, 169.9	20.5, 169.7	20.6, 169.8	20.8, 170.0
Ac (C-4g)	20.5,169.5	20.5,169.5	20.6, 169.7	20.5, 169.5	20.5, 169.5
Ac (C-5)	21.1,169.4	–	–	–	21.2, 169.6
Ac (C-7)	21.2;167.8	21.2,168.32	–		–
Ac (C′-3)	20.71, 168.1	20.7, 168.1	20.7, 168.9	20.7, 168.9	20.7, 168.9
Ac (C′-4)	20.7, 167.9	20.75,167.8			
OMe (C-6g)	52.7	52.8	52.5	52.7	52.6
OMe (C-5)	–		56.4	–	–
OMe (C-7)	–		55.8	55.8	56.0
OMe (C′-4)	–		56.0	56.0	56.0

163.1(H-1′/C-3) in the HMBC spectrum of **6** confirmed the C–C linkage between C-2 of the xanthone and C-1 of the sugar. Compound **6**, mangiferin 2′,3′,4′,5′-tetraacetate, has been prepared previously from mangiferin (Faizi et al., 2006) but the C-13 NMR spectrum is absent from the literature.

That substance **7** was the tetramethoxy derivative of **6** was obvious from the ^{1}H and ^{13}C NMR spectra (Tables 3 and 4). It has been prepared previously by partial synthesis (Aritomi and Kawasaki, 1970) although detailed spectroscopic characterization was lack. Ing, The ^{1}H and ^{13}C NMR spectra of xanthone **8** (Tables 3 and 4) exhibited a xanthone ring substitution pattern identical with that of **7** except for the substitution of one methyl by an aromatic acetyl group at δ_H 2.35 (3H, s). A cross-peak in the ROESY spectrum between the δ 2.35 signal of the acetate on C-7 and H-8 and, in the HMBC spectrum, a long range correlation between the signal of the acetyl methyl group on C-7 and the quaternary carbon at δ_C 137.2 supported the structural assignments in ring A.

In new xanthone **9** the spectroscopic evidence (Tables 3 and 4) indicated that the signals of the methyl group on C-1 of **8** were replaced by those of an acetate; moreover the interaction between the neighboring tetraacetylglucoside on C-2 and the new acetate on C-1 at room temperature resulted in formation of a rotameric

Table 3
^{1}H NMR spectra of xanthones **6**–**9**, $CDCl_3$, 600 MHz.

	6	7	8	9
H-4	6.32 s	6.69 s	6.69 s	6.79 s
H-5	6.69 s	6.83 s	6.88 s	6.88 s
H-8	8.43 s	7.64 s	7.91 s	7.84 s
C-5(OH)	13.60 brs			
H-1g	5.30 d (10.0)	5.13 d (10.0)	5.12 d (10.0)	4.80 d (10.0)
H-2g	5.53 brt (9.6)	6.03 brt (9.6)	6.02 br t 9.55)	5.92 brt (9.3)
H-3g	5.49 t (9.2)	5.34 t (9.5)	5.34 t (9.4)	5.34 t 9.6)
H-4g	5.32 t (9.66)	5.20 t (9.7)	5.20 t (9.75)	5.20 t (9.6)
H-5g	4.01 ddd (9.7, 3.8, 2.5)	3.85 ddd (9.7, 5.2, 1.8)	3.86 ddd (9.7, 5.4, 1.8)	3.80 m
H-6ag	4.36 dd (12.6, 3.8)	4.22 dd (12.3, 5.8)	4.23 dd (12.35, 5.4)	4.25 dd (12.4, 4.1)
H-6bg	4.27 dd (12.6, 2.5)	4.14 dd (12.3, 1.8)	4.15 dd (12.35, 1.8)	4.15 brd (12.4)
Ac 2g	1.94 s	1.75 s	1.78 s	1.78 s
Ac 3g	2.05 s	2.03 s	2.03 s	2.03 s
Ac 4g	2.10 s	2.05 s	2.05 s	2.07 s
Ac 6g	2.16 s	2.07 s	2.08 s	2.08 s
Ac 1				2.53 s
Ac 7			2.35 s	2.32 s
OMe (C-1)		3.94 s	3.92 s	
OMe (C-3)		4.00 s	4.01 s	4.04
OMe (C-6)		4.00 s	3.95 s	3.94 s
OMe (C-7)		3.98 s		

Table 4
^{13}C NMR of xanthones **6**–**9**, $CDCl_3$, 150 MHz.

	6	7	8	9
C-1	160.5	160.6	160.8	150.0
C-2	103.1	114.1	114.3	114.2
C-3	163.1	163.6	164.0	163.8
C-4	95.7	96.5	96.6	98.0
C-4a	157.7	159.6	159.6	158.9
C-10a	151.9	154.8	154.3	154.5
C-5	102.7	98.9	99.8	99.8
C-6	152.2	154.8	156.4	156.7
C-7	141.5	146.7	137.2	137.4
C-8	108.2	105.5	119.9	120.0
C-8a	112.9	115.6	116.0	115.5
C-9	179.7	173.9	173.4	173.4
C-9a	102.1	109.4	109.4	108.0
C-1′	73.0	72.3	72.2	72.3
C-2′	70.8	68.9	68.9	68.6
C-3′	73.5	74.9	74.9	74.5
C-4′	67.9	68.8	68.8	68.6
C-5′	76.3	76.3	76.3	76.2
C-6	61.6	62.6	62.6	62.4
OMe C-1		63.3	63.4	
OMe (C-3)		56.3	56.4	56.5
OMe (C-6)		56.3	56.5	56.4
OMe (C-7)		56.4		

Acetates: **6**: 20.75, 20.72, 20.66, 20.65; 170.9, 170.7, 170.2, 169.6; **7**: 20.5, 20.6, 20.75, 20.7; 170.7, 170.3, 169.8, 169.2; **8**: 20.77, 20.72, 20.69, 20.56, 20.52;170.72, 170.29, 169.80, 169.30, 168.9; **9**: 21.3,20.8, 20.7,x2, 20.5x2, 170.7, 170.3, 170.0, 169.7, 168.8, 168.7.

pair in the ratio 2:1 caused by restricted rotation around the linkage between the anomeric carbon and C-2 of the xanthone nucleus. Thus at room temperature in $CHCL_3$ the chemical shifts of H-3 of the two rotamers for the anomeric proton were δ_H 4.80/*5.12* (1H, d, J = 10 Hz, δ_C 72.3/*71.8*), for H-2 g, δ_H 5.92/*5.67* (1H, brt, J = 9.3 Hz, δ_C 68.6/70.6) Values in italics are for the minor conformer. When the temperature was raised the signals broadened and coalesced at 350 °C.

We conclude with some general remarks. Compounds **1** and **2** do not differ in the degree of alkylation of their quercetin hydroxyls and may well have been formed under our conditions by partial as well as complete acylation of the same naturally occurring quercetin precursor with R_1, R_2, R_3, R_4, R_5 = H and a probably unacetylated glucoyranuronate moiety. Presence of an unacetylated glucopyranuronate moiety in the naturally precursors of compounds **3**–**5** is also highly probable.

Among the four xanthone derivatives **6**–**9** failure, in the case of **6**, to undergo further acetylation under our conditions was surprising when compared with the reactivity of the two other naturally occurring precursors of xanthones **8** and **9** and since mangiferin octaacetate (R_1, R_2, R_3, R_4 = Ac) is known (Bhatia et al., 1967).

3. Experimental

3.1. General

Melting points are uncorrected and were taken on a Reichert Thermovar apparatus. Optical rotations were determined using a Perkin Elmer 2H polarimeter with a 1 dm cell. IR spectra were recorded on a Bruker TSF-55 spectropolarimeter. ^1H and ^{13}C NMR spectra were measured using Bruker Advance II 500 and Bruker Advance III 600 spectrometers. ESI-TOF and exact mass measurements were determined using a Micromass LCT Premier XE instrument. Column chromatography was performed over Amberlite XAD 2 (Supelco XAD-2-3019), Sephadex LH-20 Pharmacia (ref. 17-0090-01), silica gel (Merck 2300-400 mesh), octadecyl-functionalized silica gel (Aldrich 377635-1006) and analytical TLC Merck Kieselget 60 F254. HPLC separations were carried out on a JASCO Pu-980 series pumping system equipped with a JASCO UV-975 detector and with a Waters Kromasil Si 5 mm (10 × 250 mm) column. A Mackerey-Nagel VP 250/10 nucleodur Sphinx RP, 5 μm column was used for HPLC-RP chromatography; chromatograms were visualized under UV light at 255 and 366 nm and/or sprayed with oleum followed by heating. All the solvents were distilled before use. For acetylations dry phenolic material was dissolved in the minimum volume of pyridine. Twice the amount of acetic anhydride was added and the solution was allowed to stand overnight at ambient temperature, diluted with H_2O and extracted three times with ethyl acetate. The organic phase was evaporated at reduced pressure and the residue was purified further by HPLC (SiO_2 column) using EtOAc–hexane as eluent.

3.2. Plant material

Aerial parts of *Hypericum canariense* L. were collected in May 2007 in Breña Baja, La Palma and identified by Pedro Pérez de Paz, Botany Department, Faculty of Pharmacy, University of La Laguna, where a voucher specimen (TFC 46.334) has been deposited.

3.3. Extraction and isolation of the constituents

Fresh finely divided aerial parts of *Hypericum canariense* L., 15 kg, were extracted exhaustively with ethanol at room temperature for two weeks. The extract was filtered and concentrated to reduced pressure to give a residue (300 g) which was dissolved in 2 L of distilled water and extracted first with dichloromethane which on evaporation gave 75 g of residue and then with n-butanol which on evaporation gave 90 g of residue. The remaining aqueous layer was concentrated at reduced pressure to furnish 125 g of residue. A portion (12 g) of the latter was filtered through Amberlite XAD-2 resin. The column was first rinsed with H_2O (2 L which on evaporation yielded 6.5 g of solid residue) to remove the extremely polar components while phenolic compounds remained adsorbed on the resin and were eluted with 4 L of MeOH which furnished 3.6 g of residue. HPLC-RP chromatography of the latter using a (3:2) mixture of water–methanol as eluent at a flow rate of 1.8 mL/min, showed the presence of three major constituents with retention times of 18, 13 and 7 min in the ratio 2:1:3. Acetylation of 1.5 g of this residue with acetic anhydride (10 mL) and pyridine (5 mL) for 12 h yielded, after elimination of the solvent in vacuo, 1.8 g of crude product which was rechromatographyed over a SiO_2 column, using as eluent mixtures of hex–EtOAc and EtOAc–MeOH totalling 16 fractions, 500 mL each. Frs. 11–16, EtOAc–MeOH (9:1), furnished 68 mg of pentaacetylchlorogenic acid. Fractions 5–6, eluted with hex–EtOAc, 4:1, 200 mg, were rechromatographyed over a molecular exclusion column of Sephadex LH-20 (hex:CH_2Cl_2:MeOH; 2:1:1) using fractions of 10 mL each. Subfrs 3–6 (61 mg) after HPLC (SiO_2, EtOAc–hex 3:2, flow rate 2 mL min^{-1}) gave **2** (T_R 17 min, 3 mg) and **1** (T_R 21 min, 6.5 mg). Subfrs 20–22 furnished, after purification by crystallization from hex–EtOAc, 9 mg of mangiferin tetraacetate **6**. Subfrs 9–13 (92 mg) contained an inseparable mixture; they were therefore methylated with MeI (3 mL), K_2CO_3 (200 mg) in 5 mL of dry acetone for 7 days. The residue (95 mg) obtained after filtration and removal of solvent in vacuo was partially purified by TLC (hex–EtOAc 1:1, thrice), which yielded three bands with EtOAc. The lower band furnished 3 (8.9 mg). The middle band (26.2 mg), was purified by HPLC (SiO_2, EtOAc–hex 3:2, flow rate 2 mL min^{-1}) to give **8** (T_R 26 min, 3.5 mg), **9** (T_R 32 min, 3.9 mg) and **7** (T_R 36 min, 9.5 mg). The upper band was purified by HPLC (SiO_2, EtOAc–hex 1:1, flow 2 mL min^{-1}) to give **4** (T_R 24 min, 2.5 mg) and **5** (T_R 35 min, 5.5 mg).

3.3.1. 5,7,3′,4′-Tetraacetyl-3-(6-methyl-β-D-triacetylglucopyranurorate)-quercetin (1)

White amorphous powder from ethyl acetate–hexane; MP 197–199 °C (lit 222–224°),$[\alpha]_D^{20}$ −30° (*c* = 0.09; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 1756, 1627, 1433, 1370, 1213 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 302 (3.9), 251 (4.02) ^1H NMR Table 1, ^{13}C NMR Table 2. HRESI-TOF MS *m/z* 809.1545 (calcd for $C_{36}H_{34}O_{20}Na$, 809.1541) $[M + Na]^+$.

3.3.2. 5-Hydroxy-7,3′,4′-triacetyl-3-(6-methyl-β-D-triacetylglucopyranurorate)-quercetin (2)

White amorphous powder from ethyl acetate–hexane; MP 164–166 °C; $[\alpha]_D^{20}$ −58.3° (*c* = 0.12; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 1756, 1608, 1370, 1209, 1039 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 341 (4.01), 303 (4.15), 271 (4.24); ^1H NMR Table 1, ^{13}C NMR Table 2. HRESI-TOF MS *m/z* 767.1444 (calcd for $C_{34}H_{32}O_{19}Na$, 767.1435) $[M + Na]^+$.

3.3.3. 5,7,4′-Trimethoxy-3′-acetyl-3-(6-methyl-β-D-triacetylglucopyranurorate)-quercetin (3)

White amorphous powder from ethyl acetate–hexane; MP 140–141 °C; $[\alpha]_D^{20}$ −26.3° (*c* = 0.26; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 1759, 1636, 1614, 1372, 1218, 1041 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 324 (4.03), 263 (4.1), 243 (4.08); ^1H NMR Table 1, ^{13}C NMR Table 2. HRESI-TOF MS *m/z* 725.1703 (calcd for $C_{33}H_{34}O_{17}Na$, 725.1694) $[M + Na]^+$.

3.3.4. 5-Hydroxy-7,4′-dimethoxy-3′-acetyl-3-(6-methyl-β-D-triacetylglucopyranurorate)-quercetin (4)

White amorphous powder from ethyl acetate–hexane; MP 208–210 °C; $[\alpha]_D^{20}$ −21.2° (*c* = 0.05; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 2918, 1756, 1657, 1602, 1214, 1038, 753 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 350 (3.70), 269 (3.87), 243 (3.76); ^1H NMR Table 1, ^{13}C NMR Table 2. HRESI-TOF MS *m/z* 711.1525 (calcd for $C_{32}H_{32}O_{17}Na$, 711.1537) $[M + Na]^+$.

3.3.5. 7,4′-Dimethoxy-5,3′-diacetyl-3-(6-methyl-β-D-triacetylglucupyranurorate)-quercetin (5)

White amorphous powder from ethyl acetate–hexane; MP 157–159 °C; $[\alpha]_D^{20}$ −9.0° (*c* = 0.2; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 2922, 1757, 1630, 1217, 1038, 753 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 322 (3.66), 255 (2.62), 244 (2.64); ^1H NMR Table 1, ^{13}C NMR Table 2. HRESI-TOF MS *m/z* 753.1643 (calcd for $C_{34}H_{34}O_{18}Na$, 753.1643) $[M + Na]^+$.

3.3.6. 2-β-D-Tetraacetoxyglucopyranosyl-1,3,6,7-tetrahydroxy-9H-xanthen-9-one (6)

White amorphous powder from chloroform; MP 138–140 °C; ^1H NMR Table 3, ^{13}C NMR Table 4. HRESI-TOF MS *m/z* 613.1160 (calcd for $C_{27}H_{26}O_{15}Na$, 613.1169) $[M + Na]^+$.

3.3.7. 2-β-D-Tetraacetoxyglucopyranosyl-1,3,6,7-tetramethoxy-9H-xanthen-9-one (7)

White amorphous powder from ethyl acetate–hexane; MP 158–160 °C; $[\alpha]_D^{20}$ +13.3.0° (*c* = 0.12; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 1751, 1614, 1464, 1432, 1370, 1218, 1035 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 338 (4.07), 305 (4.33), 258 (5.07); ^1H NMR Table 3, ^{13}C NMR Table 4. HRESI-TOF MS *m/z* 669.1786 (calcd for $C_{31}H_{34}O_{15}Na$, 669.1795) $[M + Na]^+$.

3.3.8. 2-β-D-Tetraacetoxyglucopyranosyl-1,3,7-trimethoxy-7-acetyl-9H-xanthen-9-one (8)

White amorphous powder from ethyl acetate–hexane; MP 204–206 °C; $[\alpha]_D^{20}$ +31.8° (*c* = 0.06; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 1749, 1626, 1605, 1451, 1370, 1216, 1034 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 329 (3.73), 301 (4.10), 250 (4.35); ^1H NMR Table 3, ^{13}C NMR Table 4. HRESI-TOF MS *m/z* 697.1746 (calcd for $C_{32}H_{34}O_{16}Na$, 697.1745) $[M + Na]^+$.

3.3.9. 2-β-D-Tetraacetoxyglucopyranosyl-3,6-dimethoxy-1,7-diacetyl-9H-xanthen-9-one (9)

White amorphous powder from ethyl acetate–hexane; MP 128–130 °C; $[\alpha]_D^{20}$ +6.41° (*c* = 0.08; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 1750, 1620, 1454, 1369, 1215, 1108, 1035 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 328 (3.96), 302 (4.33), 269 (4.07), 253 (4.37); ^1H NMR Table 3, ^{13}C NMR Table 4. HRESI-TOF MS *m/z* 725.1698 (calcd for $C_{33}H_{34}O_{17}Na$, 725.1694) $[M + Na]^+$.

3.3.10. Pentaacetylchlorogenic acid

Although this substance was originally reported a century ago (Gorter, 1908, 1911) its physical and spectroscopic properties including the ^1H and ^{13}C NMR spectra have apparently not yet appeared in the chemical literature. Gum, $[\alpha]_D^{20}$ −13.0 (*c* = 0.59; $CHCl_3$); $IR_{\lambda max}^{NaCl}$ 1745, 1634, 1505, 1429, 1371, 1212, 1113, 1045, 755 cm^{-1}, $UV_{\lambda max}$ ($CHCl_3$) nm (log ε), 281 (4.08), 243 (3.85), HRESI-TOF MS *m/z* 587.1384 (calcd for $C_{26}H_{28}O_{14}Na$, 587.1377) $[M + Na]^+$, ^1HNMR (500 MHz, $CDCl_3$) δ: 2.67 (*brd*, *J* = 16 Hz, H-2a), 2.0 (m, H-2b), 5.54 (*td*, *J* = 10.2, 4.3 Hz, H-3), 5.12 (*dd*, *J* = 10.2, 3.3 Hz, H-4), 5.57 (*brd*, *J* = 3.3 Hz, H-5), 2.67 (*brd*, *J* = 16 Hz, H-6a), 2.41 (*brd*, *J* = 16 Hz, H-6b), 6.31 *d*, *J* = 16 Hz, H-α, 7.58 (*d*, *J* = 16 Hz, H-β), 7.36 (*d*, *J* = 1.5 Hz, H′-2), 7.21 (*d*, *J* = 8.5 Hz, H′-5), 7.39 (*dd*, *J* = 8.5, 1.5 Hz, H′-6), 2.12 (s, Ac-C-1), 1.97 (s, Ac-C-c), 2.06 (s, Ac-C-5), 2.27 (s, Ac-C′-3), 2.26 (s, Ac-C′-4); ^{13}C NMR (100 MHz, $CDCl_3$) δ 78.6 (C-1), 36.6 (C-2), 66.7 (C-3), 71.5 (C-4), 67.8(C-5), 31.6 (C-6), 182.1 (COOH), 165.4 (CO), 118.2 (C-α), 143.8 (C-β), 132.8 (C′-1), 122.8 (C′-2), 142.3 (C′-3), 143.6 (C′-4), 123.8 (C′-5), 126.5 (C′-6), 20.9, 169.9 (Ac at C-1), 20.6, 170.2 (Ac at C-4), 20.8, 169.9 (Ac at C-5), 20.47, 168.0 (Ac at C'-3)*, 20.43, 167.9 (Ac at C′-4)* (*)interchangeable.

References

Aritomi, M., Kawasaki, T., 1970. A mangiferin monomethyl ether from *Mangifera indica* L. Chem. Pharm. Bull. 18, 2224–2234.

Bhatia, V.K., Ramanathan, J.D., Seshadri, T.R., 1967. Constitution of mangiferin. Tetrahedron 23, 1363–1368.

Cardona, M.L., Pedro, J.R., Seoane, E., Vidal, R., 1985. Xanthone constituent of *Hypericum canariensis*. J. Nat. Prod. 48, 467–469.

Cardona, M.L., Fernandez, I., Pedro, J.R., Seoane, E., Vidal, R., 1986. Additional new xanthones and xanthonolignoids from *Hypericum canariensis*. J. Nat. Prod. 49, 95–100.

Cardona, M.L., Fernandez, I., Pedro, J.R., Serrano, A., 1989. A new pyranoxanthone and flavonoids from *Hypericum canariensis*. Heterocycles 29, 2297–3000 Chem. Abstr. 112, 232519 (1990).

Faizi, S., Zikr-ur-Rehman, S., Ali, M., Naz, A., 2006. Temperature and solvent dependent studies on mangiferin and complete NMR spectral assignments of its acyl and methyl derivatives. Magn. Reson. Chem. 44, 838–844.

Gorter, K., 1908. Contribution to our knowledge of coffee. Liebigs Ann. Chem. 359, 217–244.

Gorter, K., 1911. Coffee IV. Liebigs Ann. Chem. 379, 110–130.

Makovetskaya, E.Y., 2001. Research on biologically active substances of *Hypericum* species. Farmatsevitchnii Zh. (Kiev) 1, 75–80 Chem. Abstr.135, 157468 (2001).

Perez de Paz, P.L., Hernandez Padron, C.E., 1999. Plantas medicinales o útiles en la flora canaria. Aplicaciones populares. Francisco Lemus editor.

Prado, B., Rabanal, R.M., Sanchez-Mateo, C.C., 2002. Evaluation of the central properties of several *Hypericum* species from the Canary Islands. Phytother. Res. 16, 740–744.

Rabanal, R.M., Arias, A., Prado, B., Hernandez-Perez, M., Sanchez-Mateo, C.C., 2002. Antimicrobial studies on three species of *Hypericum* from the Canary Islands. J. Ethnopharmacol. 1, 287–292.

Rabanal, R.M., Bonkanka, C.X., Hernandez-Perez, M., Sanchez-Mateo, C.C., 2005. Analgesic analgesic and topical anti-inflammatory activity of *Hypericum canariense* L. and *Hypericum glandulosum* Ait. J. Ethnopharmacol. 96, 591–596.

Sanchez-Mateo, C.C., Prado, B., Rabanal, R.M., 2002. Antidepressant effect of the methanol extract of several *Hypericum* species from the Canary Islands. J. Ethnopharmacol. 79, 119.

Wagner, H., Danninger, H., Seligmann, O., Farkas, L., 1970. Erste Synthese eines natürlich vor-kommenden Flavonoid-Glucuronids (Quercetin-3β-D-glucuronid). Chem. Ber. 103, 3674–3677.