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ANTIHISTAMINIC FLAVONES AND ALIPHATIC GLYCOSIDES FROM *MENTHA SPICATA*

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Key Word Index—*Mentha spicata*; Labiatae; flavonoids; aliphatic glucosides; antiallergic; exocytosis inhibitor.

Abstract—The ethyl acetate soluble portion of the methanol extract of *Mentha spicata* var. *crispa* showed inhibitory activity on exocytosis in antigen-stimulated rat basophils. The bioassay-guided separation of the components of this fraction afforded 13 compounds. The chemical structures were elucidated to be four known flavonoids, three new and five known glucosides of lower alcohols and rosmarinic acid by means of chemical and spectroscopic evidences. Among them, three compounds showed biological activity. From the non-active 1-butanol soluble portion of the methanol extract, four new and two known structurally related glycosides were isolated and characterized.

INTRODUCTION

Mentha spicata L. var. *crispa* Benth. is cultivated as an aromatic plant. Although many terpenoids and flavonoids have been isolated from plants of this genus, few studies have been focused on their biological activity as anti-histaminic agents. This paper deals with the isolation and characterization of the constituents of this species by bioassay guided fractionation.

RESULTS AND DISCUSSION

The methanol extract of the dried leaves of *M. spicata* var. *crispa* was suspended in water, then successively partitioned with hexane, ethyl acetate and 1-butanol. Each fraction was bioassayed for inhibitory activity of exocytosis of rat basophils caused by antigen-induced stimulation. This assay is used to detect promising antiallergic reagents for type I [1]. Significant activity was observed for the ethyl acetate fraction, which was subjected to repeated silica gel and RP-18 column chromatography (CC) and preparative high-performance liquid chromatography (HPLC) to obtain 13 compounds (**1**–**5**, **7**–**10** and **15**–**18**). From the biologically non-active 1-butanol fraction, six compounds (**6**, **11**–**14** and **19**) were isolated.

Compounds **1**–**4** were identified as 5-desmethoxynobiletin [2], 5,6-dihydroxy-7,8,3′,4′-tetramethoxyflavone [2], 5,6,4′-trihydroxy-7,8,3′-trimethoxyflavone (thymonin) [3] and 5,3′,4′-trihydroxy-6,7,8-trimethoxyflavone (sideritiflavone) [2], respectively from the spectroscopic data and in agreement with those in the literature.

Compounds **5**, **7** and **8** were mono-β-glucosides based on their ^{1}H and ^{13}C NMR spectral data (Table 1). These compounds were hydrolyzed with β-glucosidase to afford the aglycones, which were identified as 1-octen-3-ol (matsutake alcohol) [4], 3-octanol and (*Z*)-3-hexenyl alcohol [5], respectively. The configuration of **5** was suggested to be 3*R*-form by comparing the spectral data in the literature [4]. The configuration of **7** was suggested to be in the 3*S*-form by comparing the ^{13}C NMR data of the glucosides with their corresponding aglycone, taking the glucosylation shift tendency into consideration [6]. From these results, **5**, **7** and **8** were identified as (3*R*)-1-octan-3-yl β-D-glucopyranoside, (3*S*)-octen-3-yl β-D-glucopyranoside and (*Z*)-3-hexenyl β-D-glucopyranoside, respectively.

Compound **6** had the molecular formula $C_{16}H_{34}O_{10}$. Its ^{13}C and ^{1}H NMR spectra showed signals for **5** and an additional pentose. The ^{13}C NMR signals of the sugar moiety suggested substitution at C-6 of the glucosyl unit, and the total sugar signals were superimposable with those of primeveroside [β-D-xylopyranosyl-(1 → 6)-β-D-glucopyranoside] reported in the literature [7]. Therefore, the structure of **6** was determined as (3*R*)-1-octen-3-yl β-D-primeveroside.

Compound **9** had the molecular formula $C_{16}H_{26}O_6$.

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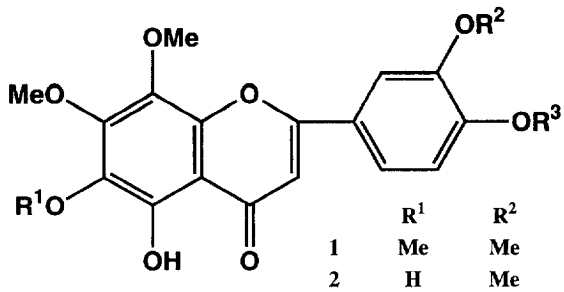
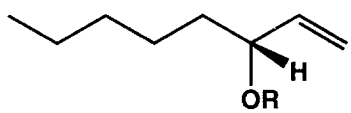
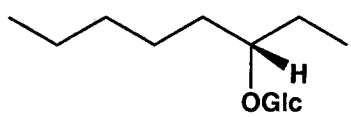


	R^1	R^2	R^3
1	Me	Me	Me
2	H	Me	Me
3	H	Me	H
4	Me	H	H



5 : R = Glc

6 : R = Glc $\overset{6}{-}$ Xyl



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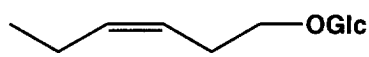

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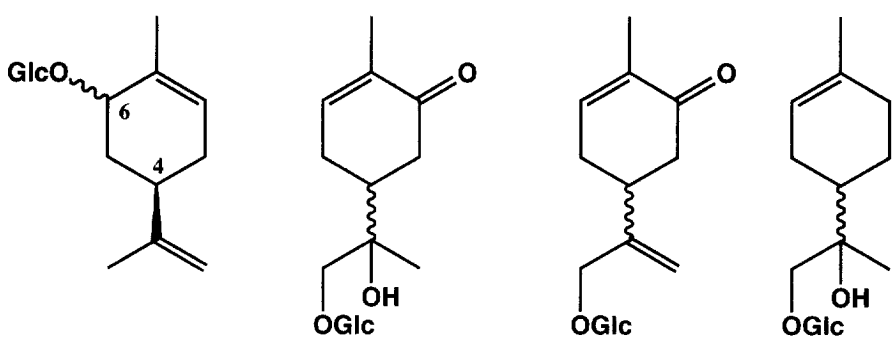


9 : 6β
10 : 6α

11

12

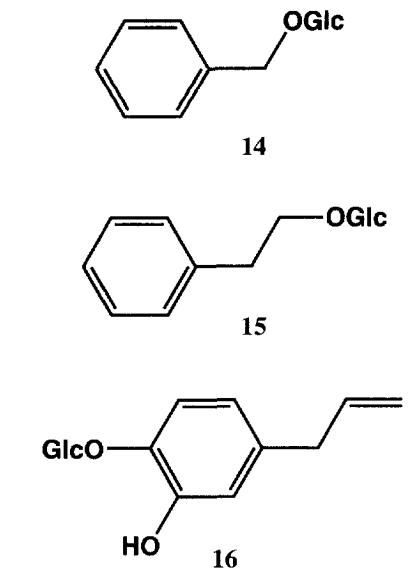
13

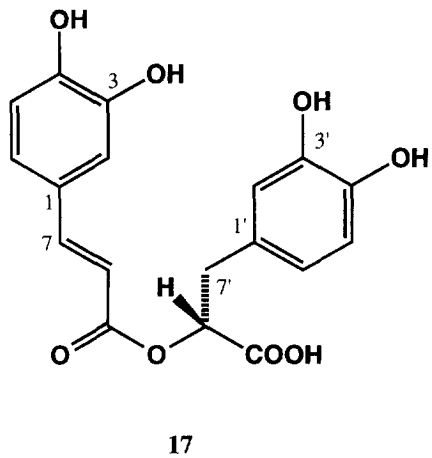
Its ^{13}C and ^{1}H NMR spectra showed signals for carveol β-D-glucopyranoside [8]. The configuration of **9** at C-6 was decided to be *S* based on comparison of the glucosylation shift of **9** with its aglycone, which was obtained by enzymatic hydrolysis of **9** with β-glucosidase. The optical rotation of the aglycone of **9** was determined to be $[\alpha]_D$ −260, which was compared with an authentic sample. From the result, the absolute structure of **9** was identified to be (4*R*, 6*S*) carveol β-D-glucopyranoside.

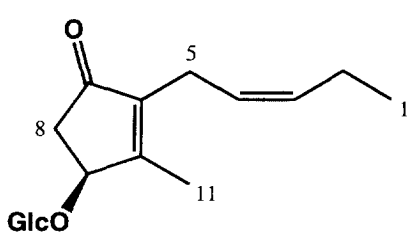
The NMR spectra of **10** were similar to that of **9**. In the same manner as **9**, the structure of **10** was decided to be (4*R*, 6*R*) carveol β-D-glucopyranoside.

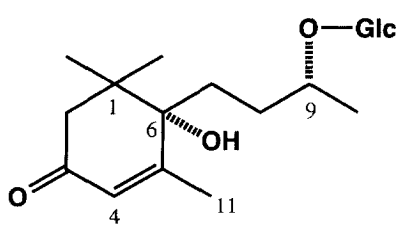
Compounds **11**–**13** had 16 signals including those of β-glucosyl carbons in their ^{13}C NMR spectrum. The ^{13}C and ^{1}H NMR spectral data of **11** indicated the presence of a trisubstituted double bond, α,β-unsaturated ketone, a quaternary methyl, a tertiary hydroxy and an O-substituted methylene group. The H-H COSY spectrum of **11** showed the partial structure: $=C=CH-CH_2-CH-CH_2-$. The chemical shift of the anomeric carbon (104.8) suggested the glucose was primary not tertiary. The structure of **11** was confirmed by comparison of the NMR data.

The NMR spectra of **12** were similar to those of **11**, but instead of the methyl and hydroxylated tertiary carbon signals, an exomethylene system was revealed. Accordingly the structure of **12** was decided as shown.









Similarly, since the NMR of **13** had a methylene signal instead of the carbonyl in **11**, the structure was decided as shown. The aglycone (or its diastereomer) was first isolated from human urine [9].

These glucosides, **11**–**13** were isolated for the first time, however, their absolute configurations could not be clarified because of the small amounts.

Compounds **14** and **15** were obtained as oil, and identified as benzylalcohol β-D-glucoside and phenethyl alcohol β-D-glucoside, respectively, by comparison of their spectral and physical data with those in the literatures [7, 10].

Compound **16** had a molecular formula of $C_{15}H_{20}O_7$. The ^{13}C and ^1H NMR spectra of **16** showed the presence of a 1,2,4-trisubstituted (two oxygen substituted) aromatic ring, an exomethylene and a glucose moiety. Based on these data, **16** was assumed to be an allylcatechol glucoside. In the NOE experiment, cross peaks were observed (i) between an anomeric proton and H-5, and (ii) between H-7 and aromatic protons, H-2 and H-6, which confirmed the location of the allyl moiety and the structure of **16** was determined as 3,4-dihydroxylallylbenzene 4-*O*-β-D-glucoside.

Compound **17** was a dimeric phenylpropanoid and was identified as rosmarinic acid, which is widely distributed in the Boraginaceae and Labiatae. This compound was reported to have anti-histaminic activity on rat peritoneal mast cells [11].

Compound **19** exhibited a quasi-molecular ion peak $[M-H]^-$ at m/z 387 in FAB MS measured in negative ion mode which corresponds to the molecular formula $C_{19}H_{12}O_8$. The ^{13}C and ^1H NMR spectra of **19** showed a double bond, a ketone group, two hydroxyl groups and a glucose moiety. The data suggested a megastigmane skeleton. The chemical shifts of the carbon signals and the optical rotation and CD data of **19** were identical with those of icariside B_5 [12]. Compound **19** was determined to be icariside B_5 as shown.

Compound **18** had a molecular formula of $C_{17}H_{26}O_7$. The ^{13}C and ^1H NMR spectra revealed the presence of a ketone group, a hydroxyl group, a *cis* double bond, a tetra-substituted double bond and a glucose moiety. The HMBC experiment showed correlations between the H-5 signal and the C-3, C-4, C-6, C-7 and C-10 signals. Also correlations were observed between the C-6 signal and the H-5, H-8, H-9 and H-11 signals. On enzymic hydrolysis with β-glucosidase, **18** gave an aglycone, **18a**, and a sugar

Table 1. ^{13}C NMR data of linear aliphatic alcohol glycosides (**5**–**8** and **18**) and the aglycone **18a**

C	**5** CD_3OD	**6** CD_3OD	**7** CD_3OD	**8** DMSO	**18** CD_3OD	**18a** CD_3OD
1	141.0	140.9	10.0	68.1	14.4	13.9
2	116.0	116.2	28.6	27.5	21.5	21.5
3	82.8	82.7	81.9	132.9	133.7	133.7
4	33.0	33.0	33.3	125.1	125.4	125.5
5	25.6	25.7	25.7	20.8	21.7	21.8
6	35.6	35.7	34.4	14.2	142.1	141.2
7	23.6	23.6	23.6		207.8	207.8
8	14.4	14.4	14.4		44.3	45.0
9					80.8	72.0
10					170.1	172.3
11					14.5	14.5
Glc 1	103.2	103.3	103.5	102.8	105.7	
2	75.3	75.3	75.3	73.4	75.2	
3	78.2	78.1	77.7	76.8	78.1	
4	71.6	71.2	71.8	70.1	71.5	
5	77.8	77.6	78.2	76.9	78.0	
6	62.8	69.5	62.9	61.1	62.7	
Xyl 1		105.3				
2		74.9				
3		77.0				
4		71.5				
5		66.8				

Table 2. ^{13}C NMR data of monoterpene glucosides (**9**–**13**)

C	**9** C_5D_5N	**10** C_5D_5N	**11** CD_3OD	**12** CD_3OD	**13** CD_3OD
1	149.6	149.3	135.9	136.1	134.8
2	125.4	124.5	147.8	147.4	121.7
3	31.4	31.4	28.7	32.6	32.0
4	36.1	41.0	42.8	43.9	41.5
5	36.0	34.8	39.6	39.3	24.1
6	78.5	76.3	203.1	202.2	28.1
7	21.4	20.4	15.6	15.7	19.5
8	134.1	136.1	73.8	148.8	78.0
9	109.1	109.3	76.2	113.4	75.1
10	20.9	20.0	20.6	71.6	23.6
Glc 1	106.7	102.0	104.8	103.2	105.1
2	75.5	75.3	75.2	75.1	75.3
3	78.1	78.6	77.9	78.2	77.1
4	71.9	72.2	71.6	71.7	71.7
5	78.1	78.4	78.0	78.0	77.9
6	63.1	63.2	62.7	62.8	62.8

which was identified as β-D-glucopyranose. The configuration at C-9 was deduced to be *S* form by comparison of the glucosylation shift (^{13}C NMR) of **18** with its aglycone (**18a**).

Each compound was bioassayed for inhibitory activity of exocytosis of rat basophils caused by antigen-induced stimulation [13]. Compound **3** showed strong activity (IC_{50} = 6.4 μM), while **2** showed mild activity (IC_{50} = 56 μM) and **5** showed weaker activity (IC_{50} = 560 μM). From the data, in case of the flavonoids, the catechol structure in B-ring is necessary to exhibit activity. The substitution pattern in the A-ring seems to have only little influence on the allergic activity of the flavonoids. This tendency is consistent with the previously reported observation [14, 15].

As far as we know, it is the first time that glucosides of volatile compounds have been found to have antihistaminic activity.

EXPERIMENTAL

General

Mps: uncorr.; ^{1}H NMR and ^{13}C NMR (TMS as int. standard): 400 and 100 MHz, respectively; FAB-MS: negative mode.

Measurement of hexosaminidase release

The secretion of β-hexosaminidase was measured as reported previously [13].

Plant material

Leaves of *Mentha spicata* L. var. *crispa* Benth. were collected in Minami-ku, Hiroshima, Japan during June 1994. A voucher specimen is deposited in the Herbarium of Institute of Pharmaceutical Sciences, Hiroshima University School of Medicine.

Extraction and isolation

Dried leaves (350 g) of *M. spicata* were extracted with hot MeOH to give 65 g of extract, a part (64 g) of which was dissolved in H_2O and then extracted with hexane, EtOAc, and *n*-BuOH, successively, to give 18 g, 7.4 g, 15 g of the respective extracts, and the remaining aq. portion (23 g). The bioactivity of each of these frs was measured at a concentration of 1mg/ml soln and found to be 100, 100, 13 and 0%, respectively. The portion of the most active fr. (EtOAc) was separated by silica gel CC using CH_2Cl_2–MeOH (30:1–5:1), CH_2Cl_2–MeOH–H_2O (15:6:1) and MeOH to give 8 frs (E-1 to E-8). The weights (mg) and activities (%) at 1 mg/ml and 100 μg/ml of E-1 to E-8 were: 67 (0 and 0), 95 (92 and 36), 490 (99 and 31), 630 (85 and 95), 1400 (98 and 51), 620 (98 and 61), 1400 (87 and 0) and 1800 (85 and 0), respectively. E-2 was fractionated on HPLC (ODS) using MeOH–H_2O (4:1) to yield **1** (12 mg). E-3 and E-4 overlapped on TLC, so these were mixed and further fractionated on HPLC(ODS) using MeOH–H_2O (3:2) to yield **2** (154 mg), **3** (130 mg) and **4** (51 mg). E-5 was further fractionated by silica gel CC using hexane–EtOAc (1:2), EtOAc–MeOH (20:1 to 1:1) and MeOH to give 6 frs (5-1 to 5-6). The weights (mg) and activities (%) at 1 mg/ml of 5-1 to 5-6 were: 46 (90), 490 (95), 300 (0), 230 (62), 150 (98) and 160 (100), respectively. Fr. 5-2 was fractionated on a column of ODS using MeOH–H_2O (3:2), and successively purified on HPLC (ODS) using MeOH–H_2O (2:3 and 1:1) to yield **5** (57.2 mg), **7** (12.8 mg), **8** (19.6 mg), **9** (57.1 mg), **10** (26.4 mg), **15** (22.3 mg), **16** (41.3 mg) and **18** (12.8 mg). From frs E-6, E-7 and E-8, **17** (210 mg) was obtained.

The *n*-BuOH extract was separated by silica gel CC using CH_2Cl_2–MeOH (5:1), CH_2Cl_2–MeOH–H_2O (15:6:1 and 6:4:1) and then MeOH to give 4 frs (B-1 to B-4). Fr. B-2 (0.69 g) was further chromatographed on a column of silica gel using EtOAc–MeOH (10:1–2:1) and MeOH to give 5 frs (B-2-1 to B-2-5). Fr. B-2-1 (470 mg) was fractionated on a column of ODS using MeOH–H_2O (1:19–1:1), and successively purified on HPLC (ODS) using MeOH–H_2O (3:17 and 3:7) and using CH_3CN–H_2O (1:9 and 3:17) to yield **12** (8.8 mg) and **14** (8.0 mg). Fr. B-2-2 (450 mg) was fractionated on a column of ODS using MeOH–H_2O (1:9–3:2), and successively purified on HPLC (ODS) using MeOH–H_2O (1:4 and 1:1) and using CH_3CN–H_2O (3:17 and 3:7) to yield **6** (7.3 mg), **13** (6.8 mg) and **19** (5.6 mg). Fr. B-2-3 (220 mg) was purified on HPLC (ODS) using MeOH–H_2O (3:7) to yield **11** (28.4 mg).

3-O-β-D-*Glucopyranosyl* (3R)-1-*octen*-3-*ol* (**5**). ^{13}C NMR: Table 1, ^{1}H NMR (CD_3OD): δ 5.19 (*d*, *J* = 16.6 Hz, H-1), 5.08 (*d*, *J* = 10.5 Hz, H-1), 5.87 (*ddd*, *J* = 16.6, 10.5, 5.9 Hz, H-2), 4.11 (*dt*, *J* = 5.9, 5.9 Hz, H-3), 1.68 (*m*, H-4a), 1.52 (*m*, H-4b), 1.31 (*m*, 6H, H-5-7), 0.89 (*t*, 3H, *J* = 6.8 Hz, H-8), 4.31 (*d*, *J* = 7.8 Hz, H-1′). FAB-MS (Neg.) *m/z*: 289 $[M-H]^-$, $[\alpha]_D^{21}$ +10.0° (MeOH; *c* 0.52).

3-O-[β-D-*Xylopyranosyl*-(1-6)-β-D-*glucopyranosyl*] (3R)-1-*octen*-3-*ol* (**6**). ^{13}C NMR: Table 1, ^{1}H NMR (CD_3OD): δ 5.10 (*ddd*, *J* = 1.2, 1.7, 17.3 Hz, H-1), 5.21 (*ddd*, *J* = 1.2, 1.7, 10.5 Hz, H-1), 5.86 (*ddd*, *J* = 6.8, 10.5, 17.3 Hz, H-2), 4.13 (*dd*, *J* = 6.8, 12.8 Hz, H-3), 1.31 (*m*, H-4), 1.52 (*m*, H-4), 1.66 (*m*, H-5, 6, 7), 0.89 (*t*, *J* = 6.8 Hz, H-8), 4.33 (*d*, *J* = 7.3 Hz, H-1′), 4.31 (*d*, *J* = 7.8 Hz, H-1″), HR-FAB-MS *m/z* 421.2114 $[M-H]^-$, $C_{19}H_{33}O_{10}$ requires 421.2074, $[\alpha]_D^{21}$ −60° (MeOH; *c* 0.13).

3-O-β-D-*Glucopyranosyl* (3S)-3-*octanol* (**7**). ^{13}C NMR: Table 1, ^{1}H NMR (CD_3OD): δ 0.91 (*t*, *J* = 7.3 Hz, H-1), 1.58 (*qd*, *J* = 7.3, 5.9 Hz, H-2), 3.64 (*tt*, *J* = 5.9, 6.4 Hz, H-3), 1.25–1.54 (*m*, H-4-7), 0.90 (*t*, *J* = 7.1 Hz, H-8), 4.30 (*d*, *J* = 7.8 Hz, H-1′), HR-FAB-MS *m/z*: 291.183 $[M-H]^-$, $C_{14}H_{27}O_6$ requires 291.1808, $[\alpha]_D^{21}$ −29° (MeOH; *c* 0.24).

β-D-*Glucopyranosyl* (Z)-3-*hexenol* (**8**). ^{13}C NMR: Table 1, ^{1}H NMR (CD_3OD): δ 2.37 (*q*, *J* = 6.8 Hz, H-2), 5.38 (*m*, H-3), 5.43 (*m*, H-4), 2.07 (*quin*, *J* = 7.1 Hz, H-5), 0.96 (*t*, *J* = 7.6 Hz, H-6), 4.26 (*d*, *J* = 7.8 Hz, H-1′), FAB-MS (Neg.) *m/z*: 261 $[M-H]^-$, $[\alpha]_D^{21}$ −30° (EtOH; *c* 0.17).

(4*R*, 6*S*)-*Carveol* β-D-*glucoside* (**9**). ^{13}C NMR: Table 2, ^{1}H NMR (C_5D_5N): δ 4.16 (*m*, H-2), 1.56 (*m*, H-3), 2.58 (*m*, H-3), 2.54 (*m*, H-4), 1.81 (*m*, H-5), 2.09 (*m*, H-5), 5.52 (*br d*, *J* = 4.4 Hz, H-6), 1.97 (*s*, H-7), 4.70 (*s*, H-9), 1.64 (*s*, H-10), 4.95 (*d*, *J* = 7.5, H-1′), FAB-MS (Neg.) *m/z*: 313 $[M-H]^-$, $[\alpha]_D^{21}$ −87.7° (MeOH: *c* 0.84).

(4*R*, 6*R*)-*Carveol* β-D-*glucoside* (**10**). ^{13}C NMR: Table 2, ^{1}H NMR (C_5D_5N): δ 4.17 (*dd*, *J* = 9.3, 8.8 Hz, H-2), 1.67 (*ddd*, *J* = 9.3, 12.2, 11.2 Hz, H-3), 2.43 (*ddd*, *J* = 8.8, 11.2, 12.2 Hz, H-3), 2.16 (*tt*, *J* = 11.2, 12.2 Hz, H-4), 1.89 (*m*, H-5), 1.95 (*m*, H-5), 5.48 (*br d*, *J* = 3.4 Hz, H-6), 1.91 (*s*, H-7), 4.72 (*m*, H-9), 1.61 (*s*, H-10), 4.97 (*d*, *J* = 7.8 Hz, H-1′), FAB-MS (Neg.) *m/z*: 313 $[M-H]^-$, $[\alpha]_D^{21}$ −40° (MeOH; c 0.35).

Compound **11**. ^{13}C NMR: Table 2, ^{1}H NMR (CD_3OD): δ 1.11 (*s*, H-10), 1.78 (*t*, J = 1.2 Hz, H-7), 6.85 (*m*, H-2), 4.26 (*d*, J = 7.8 Hz, H-1′), 3.94 (*d*, J = 10.3 Hz, H-9), 3.38 (*d*, J = 10.3 Hz, H-9), 2.61 (*dt*, J = 14.6, 2.2 Hz, H-5), 2.44 (*m*, H-3), 2.32 (*d*, J = 14.6 Hz, H-5), 2.37 (*m*, H-4), 2.22 (*m*, H-3), HR-FAB-MS *m/z*: 345.154 $[M-H]^-$, $C_{16}H_{25}O_8$ requires 345.155, $[\alpha]_D^{21}$ −7.3° (MeOH; *c* 0.55).

Compound **12**. ^{13}C NMR: Table 2, ^{1}H NMR (CD_3OD): δ 6.86 (*m*, H-2), 2.61 (*m*, H-3), 2.35 (*ddq*, J = 10.5, 18.5, 2.7 Hz, H-3), 2.93 (*m*, H-4), 2.58 (*ddd*, J = 1.5, 4.2, 16.1 Hz, H-5), 2.47 (*dd*, J = 12.7, 16.3 Hz, H-5), 1.74 (*quin*, J = 1.2 Hz, H-7), 5.20 (*s*, H-9), 5.01 (*s*, H-9), 4.41 (*d*, J = 12.4 Hz, H-10), 4.16 (*d*, J = 12.4 Hz, H-10), 4.26 (*d*, J = 7.8 Hz, H-1′), HR-FAB-MS *m/z*: 327.141 $[M-H]^-$, $C_{16}H_{23}O_7$ requires 327.1376, $[\alpha]_D^{21}$ −68° (MeOH; *c* 0.18).

*Uroterpenol β-*D*-glucoside* (**13**). ^{13}C NMR: Table 2, ^{1}H NMR (CD_3OD): δ 1.03 (*s*, H-10), 1.28 (*m*, H-4), 1.62 (*s*, H-7), 1.77 (*m*, H-6), 2.00 (*m*, H-3, 5), 3.94 (*d*, J = 10.0 Hz, H-9), 3.30 (*d*, J = 10.0 Hz, H-9), 4.25 (*d*, J = 7.8 Hz, H-1′), 5.34 (*m*, H-2), FAB-MS (Neg.) *m/z*: 331 $[M-H]^-$, $[\alpha]_D^{21}$ −38° (MeOH; *c* 0.13).

3,4-*Dihydroxyallylbenzene*-3-O-*β*-D-*glucoside* (**16**). ^{13}C NMR (C_5D_5N): δ 136.7 (C-1), 115.6 (C-2), 145.1 (C-3), 149.7 (C-4), 119.9 (C-5), 120.3 (C-6), 39.9 (C-7), 117.6 (C-8), 138.2 (C-9), 105.5 (C-1′), 79.1 (C-2′), 78.3 (C-3′), 75.0 (C-4′), 71.2 (C-5′), 62.3 (C-6′), ^{1}H NMR (C_5D_5N): δ 7.46 (*d*, J = 8.1 Hz, H-5), 7.12 (*d*, J = 2.2 Hz, H-2), 6.66 (*dd*, J = 2.2, 8.1 Hz, H-6), 5.96 (*m*, H-8), 5.06 (*ddd*, J = 17.1, 3.4, 1.5 Hz, H-9), 5.01 (*ddd*, J = 9.0, 3.4, 1.5 Hz, H-9), 3.28 (*d*, J = 6.8 Hz, H-7), 5.38 (*d*, J = 7.6 Hz, Glc-1), HR-FAB-MS *m/z*: 311.1082 $[M-H]^-$, $C_{15}H_{19}O_7$ requires 311.1131, $[\alpha]_D^{21}$ −52.6° (MeOH; *c* 0.23).

Compound **18**. ^{13}C NMR: Table 1, ^{1}H NMR (CD_3OD): δ 0.98 (*t*, J = 7.6 Hz, H-1), 2.15 (*ddd*, J = 1.5, 7.3, 7.6 Hz, H-2), 5.38 (*dtt*, J = 17.8, 7.6, 1.5 Hz, H-3), 5.20 (*dtt*, J = 17.8, 7.3, 1.5 Hz, H-4), 2.94 (*br d*, J = 7.3 Hz, H-5), 2.75 (*dd*, J = 5.9, 18.8 Hz, H-8), 2.50 (*dd*, J = 1.5, 18.8 Hz, H-8), 4.69 (*br d*, J = 5.9 Hz, H-9), 2.15 (*s*, H-11), 4.46 (*s*, J = 7.8 Hz, H-1′), HR-FAB-MS *m/z*: 341.1609 $[M-H]^-$, $C_{17}H_{25}O_5$ requires 341.1600, $[\alpha]_D^{21}$ −25° (MeOH; *c* 0.24).

Aglycone of **18** (**18a**). ^{13}C NMR: Table 1, ^{1}H NMR (CD_3OD): δ 0.97 (*t*, J = 7.6 Hz, H-1), 2.15 (*ddd*, J = 1.5, 7.3, 7.6 Hz, H-2), 5.36 (*dtt*, J = 17.8, 7.6, 1.5 Hz, H-3), 5.23 (*dtt*, J = 17.8, 7.3, 1.5 Hz, H-4), 2.93 (*br d*, J = 7.3 Hz, H-5), 2.71 (*dd*, J = 6.1, 18.5 Hz, H-8), 2.18 (overlap, H-8), 4.64 (*br d*, J = 6.1 Hz, H-9), 2.14 (*s*, H-11), FAB-MS (Neg.) *m/z* 179 $[M-H]^-$.

Icariside B_5 (**19**) [12]. $[\alpha]_D^{21}$ −20° (MeOH; *c* 0.1), {Ref. [12]: −13° (MeOH; *c* 0.62)}, CD (MeOH; *c* 0.1) [θ] (nm) +32500 (220), +2700 (327), −26,100 (251), {Ref. [12]: +42,800 (218), +5700 (324), −30,800 (250)}.

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