New Guaiane-Type Sesquiterpenoid Glycosides from *Torillis japonica* Fruit

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From the water-soluble portion of the methanolic extract of *Torillis japonica* D. C. fruit (Umbelliferae), nine new guaiane-type sesquiterpenoid glycosides, including three kessane derivatives, have been isolated. Their structures were clarified by spectral and chemical investigations.

Key words *Torillis japonica* fruit; sesquiterpenoid glycoside; guaiane; kessane; torilolone; Umbelliferae

In a previous paper,[1] we reported the isolation and characterization of a hemiterpenoid polyol and monoterpenoid glycoside from the fruit of *Torillis japonica* D. C. [Japanese name "Yabujirami" (Umbelliferae)]. In this paper, we describe nine new glycosides of guaiane-type sesquiterpenoids, which were isolated from a water-soluble fraction, as described in the Experimental section. All glycosides obtained in this paper were β-D-glucopyranosides, as evidenced from their ^{13}C-NMR data (Table 2), and by enzymatic hydrolysis to yield D-glucose. Their molecular formulae were suggested from the accurate mass number of $[M+H]^+$ or $[M+Na]^+$ ion peaks in high-resolution positive FAB-MS.

Enzymatic hydrolysis of glycoside **1** ($C_{21}H_{34}O_8$, an amorphous powder, $[\alpha]_D^{21}$ −36.4°) gave an aglycone (**1a**, $C_{15}H_{24}O_3$) and D-glucose. The aglycone was identical with torilolone, which was obtained by the alkaline hydrolysis of torilin.[2] The position of the glucosyl unit was deduced to be C-11 from the observed three bond correlation between the anomeric proton and the C-11 carbon signal in the heteronuclear multiple-bond correlation (HMBC) spectrum. Therefore, **1** could be represented as torilolone 11-*O*-β-D-glucopyranoside.

Glycoside **2** ($C_{21}H_{32}O_8$, an amorphous powder, $[\alpha]_D^{21}$ +5.1°) was revealed to be an 8-oxo derivative of **1** by comparison of its ^{1}H- and ^{13}C-NMR data with those of **1** (Tables 1, 2). This was confirmed by enzymatic hydrolysis which gave aglycone **2a** ($C_{15}H_{22}O_3$), which was identical with the CrO_3 oxidation product of **1a**. Therefore, **2** was characterized as (1*R*,7*R*,10*S*)-11-hydroxyguai-4-ene-3,8-dione β-D-glucopyranoside.

Glycosides **3** ($C_{23}H_{36}O_9$, mp 228—230°C, $[\alpha]_D^{21}$ −18.3°) and **4** ($C_{23}H_{36}O_9$, an amorphous powder, $[\alpha]_D^{21}$ +22.0°) showed, in addition to a glucopyranoside moiety, three *tert*-methyls, one *sec*-methyl, three methylenes, four methines (one of them oxygenated), one tetrasubstituted double bond, one carbonyl function, one acetoxy group, and one oxygenated quaternary carbon, in their ^{1}H- and ^{13}C-NMR data (Tables 1, 2). These data and the results of HMBC experiments suggested that they had planar structures analogous to **1**. Alkaline hydrolysis of **3** using 7% NH_4OH–MeOH gave a deacetate derivative (**5**, $C_{21}H_{34}O_8$), and enzymatic hydrolysis of **5** gave an aglycone (**5a**, $C_{15}H_{24}O_3$) and D-glucose. Aglycone **5a** was not identical with **1a**, but the diketone derivatives obtained by CrO_3 oxidation of **1a** and **5a** were identical. Thus, **5a** was suggested to be a 8-epimer of **1a**. This suggestion was supported by the downfield shifts of H-7α (by 0.40 ppm) and H-10α (by 0.75 ppm), and by the upfield shift of the H_3-12 signal (by 0.17 ppm) of **5a**, when compared with those of **1a**. The position of the glucosyl and acetoxy units in **3** were ascertained to be C-8 and C-11, respectively, by the observed three bond correlation between the anomeric proton and the C-8 carbon in the HMBC spectrum, and the downfield shift of the C-11 signal (by 11.6 ppm), when compared with **5a**. The absolute configuration at C-8 was deduced to be *S* by the glycosylation shift ($\Delta\delta$ **3**—**5a**: 9.72 ppm) and the chemical shift of the glucosyl C-1 signal (δ 104.96).[3] Thus, **3** was characterized as 11-*O*-acetyl-8-*epi*-torilolone 8-*O*-β-D-glucopyranoside. Glycoside **4** was a stereoisomer of **3**. Nuclear Overhauser effect (NOE) interactions between the proton signals shown in Fig. 1 were observed in the nuclear Overhauser and exchange spectroscopy (NOESY) spectrum of **4**, and the configurations at H-1, H-7 and H-10 were concluded to be α, while H-8 was β. Further, the absolute configuration at C-8 was suggested to be *S* by comparison of the chemical shifts of the C-8 and glucosyl C-1 carbons with those of **3** [C-8 (**3**: δ 79.95, **4**: δ 81.48), glucosyl C-1 (**3**: δ 104.96, **4**: δ 104.77)]. Therefore, **4** was characterized as (1*S*,7*R*,8*S*,10*S*)-11-acetoxy-8-hydroxyguai-4-en-3-one 8-*O*-β-D-glucopyranoside.

Glycoside **5** ($C_{21}H_{34}O_8$, an amorphous powder, $[\alpha]_D^{21}$ −5.1°) was identical with the material obtained by alkaline hydrolysis of **3**. Therefore, **5** was identified as 8-*epi*-torilolone 8-*O*-β-D-glucopyranoside.

Glycoside **6** ($C_{21}H_{36}O_9$, mp 228—230 °C, $[\alpha]_D^{21}$ +6.7°) showed the presence of three *tert*-methyls, one *sec*-methyl, three methylenes, five methines (two of them oxygenated) and three oxygenated quaternary carbons in the aglycone moiety by ^{1}H- and ^{13}C-NMR data (Tables 1, 2). Enzymatic hydrolysis gave an aglycone (**6a**, $C_{15}H_{26}O_4$) and D-glucose. From analysis of the HMBC and ^{1}H–^{1}H correlated spectroscopy (COSY) spectral data for **6a**, a gross planar structure was obtained and was suggested to be a guaiane-type sesquiterpenoid with oxygenated functions at C-2, C-7, C-8, C-10 and C-11. From the molecular formula, **6a** was suggested to have a kessane skeleton having an ether ring between C-10 and C-11.[4] This was also supported by the observed NOE interactions between the proton signals shown in Fig. 1 in the NOESY spectra of **6a** and **6a**-diacetate (**6a′**). The configurations at H-2 and H-8 were indicated to be β and α, respectively, from the observed NOE interactions between H-2 and H_3-14, H-2 and H-5β, H-1α and H-8, and between H-6α and H-8. Thus, **6a** was characterized as

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Table 1. ^{1}H-NMR Chemical Shifts of **1**—**9**, **1a**, **2a** and **5a**—**7a** [in Pyridine-d_5 Except **2a** (in $CDCl_3$), 500 MHz]

	1	**1a**	**2**	**2a**
H-1	2.24 m	2.24 m	2.35 br dd (13.5, 7.0)	2.48 br dd (11.0, 6.5)
H-2α	2.09 dd (18.0, 3.5)	2.06 dd (18.0, 3.5)	2.03 dd (18.0, 1.5)	2.08 dd (18.5, 1.5)
2β	2.56 dd (18.0, 6.0)	2.59 dd (18.0, 6.0)	2.54 dd (18.0, 7.0)	2.63 dd (18.5, 6.5)
H-6α	3.19 br d (13.0)	3.09 br d (13.5)	3.20 dd (13.0, 5.0)	3.07 dd (12.0, 4.0)
6β	2.67 dd (13.0, 10.0)	2.64 dd (13.5, 9.5)	2.95 dd (13.0, 13.0)	2.56 dd (13.0, 13.0)
H-7	1.67 br dd (10.0, 4.0)	1.42 br dd (9.5, 4.5)	2.73 dd (13.0, 5.0)	2.35 dd (13.0, 4.0)
H-8	4.49 ddd (10.0, 7.5, 4.0)	4.53 ddd (10.0, 8.0, 4.0)		
H-9α	2.18 br dd (14.0, 7.5)	2.16 dd (14.0, 8.0)	3.35[a] dd (12.0, 12.0)	2.99[a] dd (12.0, 12.0)
9β	1.89 m	1.95 m	2.29[a] dd (12.0, 2.0)	2.36[a] dd (12.0, 2.0)
H-10	1.24 m	1.20 m	1.36 m	1.47 m
H_3-12	1.78 s	1.66 s	1.63 s	1.30[b] s
H_3-13	1.69 s	1.50 s	1.64 s	1.33[b] s
H_3-14	0.89 d (6.5)	0.91 d (6.5)	0.91 d (6.5)	1.15 d (6.5)
H_3-15	1.79 d (1.5)	1.82 d (1.5)	1.69 s	1.70 s
Glc-1	5.17 d (7.5)		5.08 d (8.0)	
OAc				

	3	**4**	**5**	**5a**
H-1	2.21 m	2.52 m	2.20 m	2.21 m
H-2α	1.94 dd (18.0, 2.0)	2.54 dd (17.0, 6.0)	1.94 dd (18.0, 2.0)	2.10 dd (18.5, 3.0)
2β	2.55 dd (18.0, 6.0)	2.03 br d (17.0)	2.54 dd (18.0, 6.5)	2.61 dd (18.5, 7.5)
H-6α	2.85 br dd (12.5, 2.5)	2.74 dd (17.0, 5.0)	2.82 br d (13.0)	2.79 br d (12.5)
6β	2.13 dd (13.5, 12.5)	2.69 dd (17.0, 8.0)	2.10 dd (13.0, 13.0)	2.01 dd (12.5, 11.5)
H-7	2.43 ddd (13.5, 4.0, 2.5)	2.90 ddd (8.0, 5.0, 5.0)	2.03 br dd (13.0, 4.5)	1.82 dd (11.5, 7.5)
H-8	4.22 br dd (5.0, 4.0)	4.26 br dd (8.0, 4.0)	4.45 br dd (4.5, 3.5)	4.43 ddd (7.5, 3.5, 3.5)
H-9α	2.53 br dd (14.5, 5.0)	2.36 ddd (14.5, 4.0, 4.0)	2.47 dd (14.5, 4.5)	2.02 dd (13.0, 3.5)
9β	1.79 m	2.30 dd (14.5, 7.0)	1.85 m	1.97 dd (13.0, 3.5)
H-10	2.00 m	1.43 m	1.94 m	1.95 m
H_3-12	1.89 s	1.86 s	1.46 s	1.49 s
H_3-13	1.64 s	1.71 s	1.52 s	1.53 s
H_3-14	1.09 d (6.5)	1.17 d (6.5)	1.05 d (6.5)	0.96 d (6.5)
H_3-15	1.78 d (1.5)	1.86 s	1.76 d (1.5)	1.80 d (1.5)
Glc-1	4.86 d (7.5)	5.20 d (7.5)	4.94 d (8.0)	
OAc	2.06 s	2.00 s		

2α,7,8β-trihydroxykessane. The position of the glucosyl unit in **6** was ascertained to be C-2 from the observed three bond correlation between the anomeric proton and the C-2 carbon signal in the HMBC spectrum, and the absolute configuration at C-2 was suggested to be *S* for the same reasons described for **3** [C-2 (δ 83.11, $\Delta\delta$ **6**—**6a**: 10.23 ppm), glucosyl C-1 (δ 105.54)]. Thus, **6a** was confirmed to have the same absolute configuration as kessane.[5] From these facts, **6** could be represented as 2α,7,8β-trihydroxykessane 2-*O*-β-D-glucopyranoside.

Glycosides **7** ($C_{21}H_{36}O_8$, an amorphous powder, $[\alpha]_D^{21}$ +17.6°) and **8** ($C_{21}H_{36}O_9$, an amorphous powder, $[\alpha]_D^{23}$ +10.0°) were revealed to be derivatives of **6** by comparison of ^{1}H- and ^{13}C-NMR data (Tables 1, 2), and HMBC experiments. Enzymatic hydrolysis of **7** gave an aglycone (**7a**, $C_{15}H_{26}O_3$) and D-glucose. Comparison of the ^{13}C-NMR data for **6a** and **7a** suggested that **7a** was an 8-dehydroxyl derivative of **6a**. The observed NOE interactions, shown in Fig. 1, in the NOESY spectrum of **7a** supported this conclusion. In glycoside **8**, the presence of a hydroxymethyl group was shown by the ^{1}H- and ^{13}C-NMR data. Further, **8** was indicated to be the 12-hydroxy derivative of **7** by comparison of ^{13}C-NMR data, and the observed NOE interactions between H-5β and H_3-13, and between H-6β and H_3-13 (Fig. 1) in the NOESY spectrum. As the absolute configurations at C-2 of **7** and **8** were indicated to be *S* [C-2 (**7**: δ 83.44, $\Delta\delta$ **7**—**7a**: 10.39 ppm, **8**: δ 83.38), glucosyl C-1 (**7**: δ 105.68, **8**: δ 105.65)], they were revealed to have the same absolute configuration as **6**. Thus, **7** and **8** were characterized as 2α,7-dihydroxykessane 2-*O*-β-D-glucopyranoside and 2α,7,12-trihydroxykessane 2-*O*-β-D-glucopyranoside, respectively.

Glycoside **9** ($C_{21}H_{38}O_8$, an amorphous powder, $[\alpha]_D^{21}$ +9.8°) showed the presence of three *tert*-methyls, six methylenes (one of them oxygenated), four methines (one of them oxygenated) and two oxygenated quaternary carbons, in the ^{1}H- and ^{13}C-NMR data (Tables 1, 2). From analysis of the HMBC and ^{1}H–^{1}H COSY spectral data, the aglycone of **9** was suggested to be a guaiane-type sesquiterpenoid having three hydroxyl groups at C-10, C-11 and C-15. The position of the glucosyl unit was indicated to be C-2 from the observed three bond correlation between the anomeric proton and C-2 carbon signals. The configurations at C-1, C-4, C-5, C-7 and C-10 were deduced to be the same as kessane from the observed NOE interactions in its NOESY spectrum, as shown in Fig. 1. Though the absolute configuration of **9** could not be deduced from the above data, it can be drawn as shown in Fig. 1 from a biogenetic point of view. From these results, **9** was concluded to be (1*R*,4*R*,5*R*,7*R*,10*S*)-10,11,15-trihydroxyguaiane 11-*O*-β-D-glucopyranoside.

Glycosides **6**—**8** have aglycones having structures that are

Table 1. Continued

	6	6a	7
H-1	2.20 dd (14.0, 9.0)	2.14 dd (13.0, 9.0)	2.23 dd (13.5, 9.0)
H-2α			
2β	4.11 ddd (9.0, 9.0, 5.0)	4.35 ddd (9.0, 9.0, 6.0)	4.15 ddd (9.0, 9.0, 5.0)
H-3α	1.76 br dd (14.0, 5.0)	1.44 ddd (14.0, 6.0, 2.5)	1.80 ddd (15.0, 5.0, 2.5)
3β	2.58 ddd (14.0, 9.0, 9.0)	2.52 ddd (14.0, 9.0, 9.0)	2.61 ddd (15.0, 9.0, 9.0)
H-4	1.90 m	2.00 m	1.91 bd dd (15.0, 13.0)
H-5	2.39 br ddd (14.0, 13.0, 9.0)	2.53 m	2.41 ddd (13.0, 13.0, 7.0)
H-6α	1.79 bd d (13.0)	1.87 dd (13.5, 13.5)	1.92 br d (13.0)
6β	2.30 dd (13.0, 7.0)	2.38 dd (13.5, 6.5)	2.25 dd (13.0, 13.0)
H-8α	4.21 m	4.36 dd (9.0, 8.0)	2.02 m
8β			2.27 br dd (13.0, 5.0)
H-9α	2.35 br d (2H, 8.5)	2.61 dd (13.5, 9.0)	1.78 ddd (13.0, 7.0, 5.0)
9β		2.48 dd (13.5, 8.0)	2.03 m
H_3- or H_2-12	1.68 s	1.74 s	1.56 s
H_3-13	1.54 s	1.57 s	1.55 s
H_3-14	1.77 s	1.76 s	1.75 s
H_3- or H_2-15	0.84 d (7.0)	0.95 d (7.5)	0.90 d (7.0)
Glc-1	4.79 d (6.5)		4.82 d (8.0)

	7a	8	9
H-1	2.16 dd (14.0, 8.5)	2.23 dd (13.0, 9.0)	1.79 br dd (9.0, 9.0)
H-2α			1.96 m
2β	4.37 ddd (9.0, 8.5, 6.0)	4.16 ddd (9.0, 9.0, 5.0)	1.81 br dd (9.0, 9.0)
H-3α	1.49 ddd (13.5, 5.5, 2.5)	1.80 ddd (15.0, 5.0, 2.5)	1.71 br ddd (12.0, 12.0, 4.0)
3β	2.55 ddd (14.0, 9.0,9.0)	2.61 ddd (15.0, 9.0, 9.0)	1.97 m
H-4	2.03 m	1.93 br dd (9.0, 6.5)	2.52 m
H-5	2.53 br ddd (14.0, 13.0, 7.0)	2.44 ddd (19.5, 13.5, 6.5)	2.95 dddd (9.0, 9.0, 8.0, 5.0)
H-6α	1.99 dd (13.0, 13.0)	1.88 br d (13.5)	1.86 m
6β	2.31 dd (13.0, 6.5)	2.31 dd (19.5, 13.5)	2.01 m
H-7			2.50 br t (9.0)
H-8α	2.17 m	2.10 dd (12.0, 9.5)	1.97 ddd (12.0, 12.0, 9.0)
8β	2.32 m	2.30 dd (12.0, 9.5)	2.14 m
H-9α	1.82 ddd (10.0, 10.0, 4.0)	1.59 br dd (13.0, 9.5)	1.68 br dd (12.0, 12.0)
9β	2.13 m	2.03 br dd (13.0, 9.5)	2.12 br d (12.0)
H_3- or H_2-12	1.60 s	3.90 br d (10.5)	1.38 s
		4.29 br d (10.5)	
H_3-13	1.57 s	1.78 s	1.48 s
H_3-14	1.73 s	1.73 s	1.37 s
H_3- or H_2-15	1.01 d (7.0)	0.90 d (7.5)	3.87 dd (10.5, 8.0)
			4.10 dd (10.5, 6.0)
Glc-1		4.82 d (7.5)	5.08 d (8.0)

δ in ppm from TMS [coupling constants (J) in Hz are given in the parentheses]. *a,b*) Assignments may be interchanged.

analagous to α-kessyl alcohol, which has strong antidepressant activity in mice.[6]

Experimental

The instruments and experimental conditions for obtaining spectral data and for chromatography were the same as in a previous paper.[1]

Extraction and Isolation of Guaiane-Type Sesquiterpenoid Glycosides The methanolic extract of the fruit of *Torillis japonica* D. C. was treated as described in a previous paper,[1] and the methanol eluate (11.1 g) was obtained from the aqueous portion (30.6 g) by Amberlite XAD-II chromatography. This fraction was chromatographed on Sephadex LH-20 (MeOH) to furnish five fractions (frs. 1—5). Fraction 2 (9.3 g) was purified by silica gel [$CHCl_3$–MeOH–H_2O (4 : 1 : 0.1→7 : 3 : 0.5→MeOH)] chromatography to afford seven fractions (frs. 2-1—2-7). From fr. 2-1, **3** (130 mg) and **4** (70 mg) were isolated by repeated Sephadex LH-20 (MeOH), Lobar RP-8 column [MeOH–H_2O (1 : 1)] and silica gel [$CHCl_3$–MeOH (9 : 1)] chromatographies. From fr. 2-2, **1** (18 mg), **2** (18 mg) and **5** (12 mg) were isolated by repeated Sephadex LH-20 (MeOH), Lobar RP-8 column [MeOH–H_2O (3 : 7)] chromatographies and HPLC [octadecyl silica (ODS), CH_3CN–H_2O (3 : 17 and 1 : 5)]. From fr. 2-3, **7** (159 mg) was isolated by repeated Sephadex LH-20 (MeOH), Lobar RP-8 column [MeOH–H_2O (3 : 7)] and silica gel [$CHCl_3$–MeOH–H_2O (4 : 1 : 0.1)] chromatographies. From fr. 2-4, **6** (181 mg) was isolated by Sephadex LH-20 (MeOH), Lobar RP-8 column [MeOH–H_2O (3 : 7)] and silica gel [$CHCl_3$–MeOH–H_2O (4 : 1 : 0.1)] chromatographies. From fr. 2-5, **8** (10 mg) and **9** (13 mg) were isolated by repeated Sephadex LH-20 (MeOH), Lobar RP-8 column [MeOH–H_2O (3 : 7)], silica gel [$CHCl_3$–MeOH–H_2O (3 : 1 : 0.1) for **8**; $CHCl_3$–MeOH–H_2O (4 : 1 : 0.1) for **9**] chromatographies and HPLC [ODS, CH_3CN–H_2O (3 : 37) for **8**; ODS, CH_3CN–H_2O (1 : 9) for **9**].

Torilolone 11-*O*-β-D-Glucopyranoside (1) An amorphous powder, $[\alpha]_D^{21}$ −36.4° (c=0.9, MeOH). Positive FAB-MS m/z: 415.2310 $[M+H]^+$ (base, Calcd for $C_{21}H_{35}O_8$: 415.2332), 235 $[M-C_6H_{12}O_6+H]^+$.

Enzymatic Hydrolysis of 1 A mixture of **1** (10 mg) and hesperidinase (3 mg) in water (5 ml) was shaken on a water bath at 37 °C for two weeks. The mixture was evaporated *in vacuo* to dryness and the residue was chromatographed on silica gel [$CHCl_3$–MeOH (9 : 1) and $CHCl_3$–MeOH–H_2O (7 : 3 : 0.5)] to give torilolone (**1a**; 4 mg) and the sugar fraction. The sugar fraction was passed through Sephadex LH-20 (MeOH) to give a syrup. This was analyzed by HPLC [column; carbohydrate analysis (Waters: size, 3.9× 300 mm), detector; JASCO RI-930 and OR-990 chiral detector: CH_3CN–H_2O (17 : 3), 2 ml/min; t_R 4.53 min] which revealed the presence of D-glucose.

Torilolone (1a) Colorless needles (aq. MeOH), mp 180—182 °C, $[\alpha]_D^{21}$

Table 2. ^{13}C-NMR Chemical Shifts of **1**—**9**, **1a**, **2a** and **5a**—**7a** [in Pyridine-d_5 Except **2a** (in $CDCl_3$), 125 MHz]

	1	1a	2	2a	3	4	5	5a	6	6a	7	7a	8	9
Aglycone C-1	51.84	51.89	50.17	50.17	50.11	48.34	50.23	50.89	54.66	56.32	55.24	56.97	55.31	54.44
C-2	41.57	41.57	41.82	41.50	41.84	41.99	41.71	41.64	83.11	72.85	83.44	73.07	83.38	28.97
C-3	207.83	207.79	207.11	207.61	207.90	207.44	207.87	207.66	42.50	44.13	42.74	44.36	42.63	29.65
C-4	134.21	133.85	138.37	138.36	135.89	136.20	135.30	134.76	32.23	31.76	32.41	31.88	32.39	47.31
C-5	176.60	176.78	172.43	171.30	175.43	174.98	176.32	176.27	38.18	38.83	38.50	39.11	38.12	36.40
C-6	24.75	24.83	27.15	27.63	28.12	27.83	29.28	28.77	40.18	40.43	42.74	43.00	43.02	27.49
C-7	50.84	49.50	60.51	59.49	52.39	51.81	54.19	54.98	71.68	71.79	71.53	71.59	72.61	47.49
C-8	68.84	69.90	213.13	214.54	79.95	81.48	81.45	70.23	71.93	72.10	33.57	33.70	34.06	21.67
C-9	44.65	44.57	49.83	50.12	39.48	40.07	39.92	43.26	47.79	48.37	36.91	37.41	37.21	46.47
C-10	33.44	33.60	41.56	39.86	33.30	37.54	33.11	32.31	72.89	72.95	73.94	73.95	74.37	71.22
C-11	80.53	73.05	78.42	72.22	84.92	85.00	72.64	73.29	78.38	78.40	78.61	78.59	78.75	81.45
C-12	24.41	28.91	25.06	27.23	22.90	24.16	26.08	26.01	25.80	25.89	24.87	24.92	69.93	22.83
C-13	26.89	29.51	25.68	28.62	24.57	25.76	29.61	29.41	28.32	28.18	27.77	27.84	20.33	24.52
C-14	23.09	23.06	22.60	22.83	23.02	23.29	23.15	23.44	28.56	28.48	28.67	28.24	28.32	31.04
C-15	8.25	8.13	7.78	7.77	7.76	8.65	7.88	8.02	18.21	18.71	18.20	18.75	18.11	62.86
Glucose C-1	98.17		98.57		104.96	104.77	105.23		105.54		105.68		105.65	98.56
C-2	75.21		75.27		75.12	75.63	75.40		75.40		75.49		75.49	75.40
C-3	79.07		78.24		78.30	78.36	78.42		78.07		78.16		78.20	78.08
C-4	71.98		71.81		71.50	71.97	71.55		71.83		71.92		71.92	71.92
C-5	78.38		78.88		78.65	78.67	78.58		78.52		78.61		78.62	78.69
C-6	63.02		62.89		62.88	63.12	62.85		62.98		63.05		63.07	63.00
OAc					22.56, 170.19	22.52, 170.17								

δ in ppm from TMS.

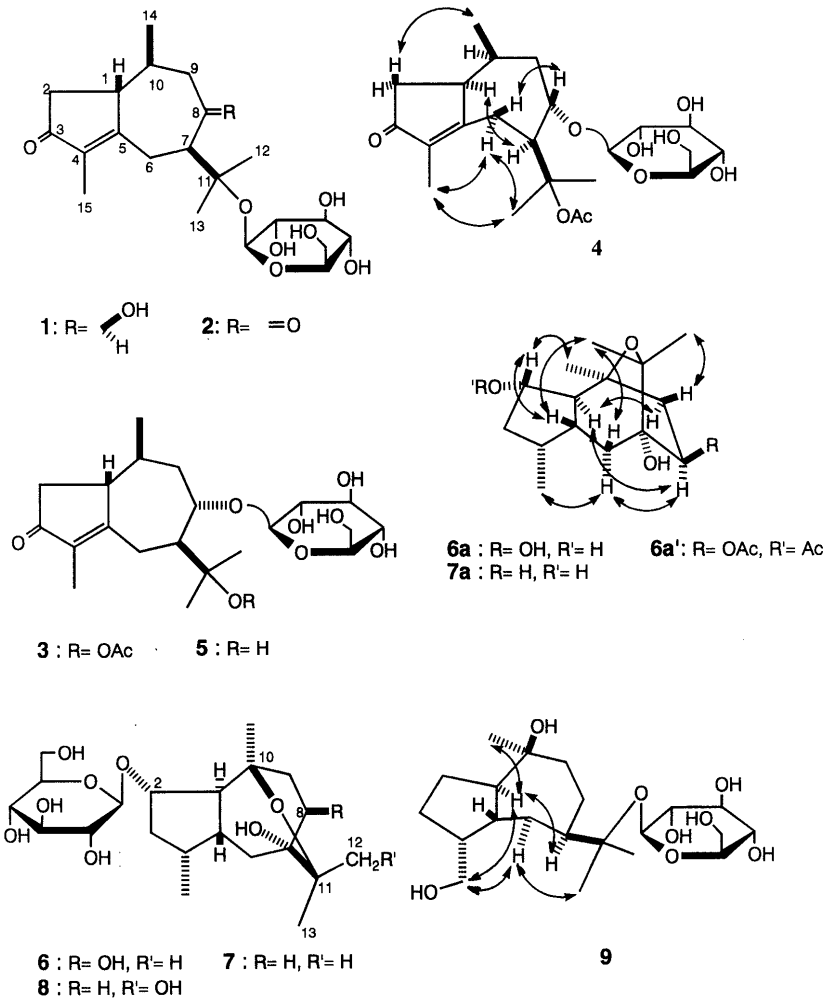

Fig. 1. Structures of **1**—**9** and NOE Interactions Observed in the NOESY Spectra of **4**, **6a**, **6a'**,**7a** and **9**

$-15.7°$ (c=0.3, MeOH) [lit.[2] mp 179—180 °C, $[\alpha]_D^8$ $-25.7°$ (c=1.83, $CHCl_3$)].

(1*R*,7*R*,10*S*)-11-Hydroxyguai-4-ene-3,8-dione β-D-Glucopyranoside (2) Amorphous powder, $[\alpha]_D^{21}$ $+5.1°$ (c=0.7, MeOH). Positive FAB-MS *m/z*: 435 $[M+Na]^+$, 413.2189 $[M+H]^+$ (base, Calcd for $C_{21}H_{33}O_8$: 413.2176), 233 $[M-C_6H_{12}O_6+H]^+$.

Enzymatic Hydrolysis of 2 A mixture of **2** (10 mg) was treated as described for **1**, and the residue was chromatographed on silica gel [hexane–EtOAc (7 : 3) and $CHCl_3$–MeOH–H_2O (7 : 3 : 0.5)] to give **2a** (4 mg) and the sugar fraction. From the sugar fraction, the presence of D-glucose was demonstrated as for **1**.

(1*R*,7*R*,10*S*)-11-Hydroxyguai-4-ene-3,8-dione (2a) An amorphous powder, $[\alpha]_D^{21}$ $+20.8°$ (c=0.3, MeOH). Positive FAB-MS *m/z*: 252 $[M+H]^+$ (base; $C_{15}H_{23}O_3$).

CrO_3 Oxidation of 1a A mixture of torilolone (20 mg), acetic acid (5 ml) and CrO_3 (5 mg) was stirred for 3 h at room temperature. The reaction mixture was then poured into water and extracted with Et_2O. The organic layer was concentrated to give a residue, which was purified by silica gel [hexane–EtOAc (7 : 3)] chromatography and HPLC [ODS, CH_3CN-H_2O (9 : 1)] to give **2a** (5.5 mg).

11-*O*-Acetyl-8-*epi*-torilolone 8-*O*-β-D-Glucopyranoside (3) Colorless needles (aq. MeOH), mp 228—230 °C, $[\alpha]_D^{21}$ $-18.3°$ (c=1.5, MeOH). Positive FAB-MS *m/z*: 913 $[2M+H]^+$, 457.2466 $[M+H]^+$ (Calcd for $C_{23}H_{37}O_9$: 457.2437), 397 $[M-CH_3COOH+H]^+$, 295 $[M-C_6H_{10}O_5+H]^+$ (base), 277 $[M-C_6H_{12}O_6+H]^+$, 235 $[M-CH_3COOH-C_6H_{10}O_5+H]^+$.

Deacetylation of 3 **3** (30 mg) was refluxed in 7% NH_4OH–MeOH on a hot water bath for 21 h, then the reaction mixture was evaporated *in vacuo* to dryness and the residue chromatographed on silica gel [$CHCl_3$–MeOH–H_2O (4 : 1 : 0.1)] and Sephadex LH-20 (MeOH) to give **5** (20 mg).

Enzymatic Hydrolysis of 5 A mixture of **5** (19 mg) and hesperidinase (5 mg) in water (5 ml) was shaken on a water bath at 37 °C for two weeks. The mixture was evaporated *in vacuo* to dryness and the residue was chromatographed on silica gel [hexane–EtOAc (3 : 2) and $CHCl_3$–MeOH–H_2O (7 : 3 : 0.5)] to give **3a** (8 mg) and the sugar fraction. From the sugar fraction, the presence of D-glucose was demonstrated as for **1**.

8-*epi*-Torilolone (3a) An amorphous powder, $[\alpha]_D^{23}$ $-27.7°$ (c=0.7, MeOH). Positive FAB-MS *m/z*: 253 $[M+H]^+$ (base; $C_{15}H_{25}O_3$).

CrO_3 Oxidation of 3a A mixture of **3a** (8 mg), acetic acid (2 ml) and CrO_3 (2 mg) was treated as described for **1a**, and the residue was purified by silica gel [hexane–EtOAc (7 : 3)] chromatography and HPLC [ODS, CH_3CN–H_2O (9 : 1)] to give **2a** (2.0 mg).

(1*S*,7*R*,8*S*,10*S*)-11-Acetoxy-8-hydroxyguai-4-en-3-one 8-*O*-β-D-Glucopyranoside (4) An amorphous powder, $[\alpha]_D^{23}$ $+22.0°$ (c=1.0, MeOH). Positive FAB-MS *m/z*: 457.2458 $[M+H]^+$ (Calcd for $C_{23}H_{37}O_9$: 457.2437), 397 $[M-CH_3COOH+H]^+$, 295 $[M-C_6H_{10}O_5+H]^+$ (base), 277 $[M-C_6H_{12}O_6+H]^+$, 235 $[M-CH_3COOH-C_6H_{10}O_5+H]^+$.

8-*epi*-Trilolone 8-*O*-β-D-Glucopyranoside (5) An amorphous powder, $[\alpha]_D^{21}$ $-5.1°$ (c=0.7, MeOH). Positive FAB-MS *m/z*: 437 $[M+Na]^+$, 415.2317 $[M+H]^+$ (base, Calcd for $C_{21}H_{35}O_8$: 415.2332), 235 $[M-C_6H_{10}O_5+H]^+$.

2α,7,8β-Trihydroxykessane 2-*O*-β-D-Glucopyranoside (6) Colorless needles (aq. MeOH), mp 228—230 °C, $[\alpha]_D^{21}$ $+6.7°$ (c=2.0, MeOH). Positive FAB-MS *m/z*: 866 $2[M+H]^+$, 433.2421 $[M+H]^+$ (base, Calcd for $C_{21}H_{37}O_9$: 433.2438), 415 $[M-H_2O+H]^+$, 379 $[M-2H_2O+H]^+$, 253 $[M-C_6H_{12}O_6+H]^+$. Negative FAB-MS *m/z*: 431 $[M-H]^-$ (base).

Enzymatic Hydrolysis of 6 A mixture of **6** (50 mg) and hesperidinase (5 mg) in water (5 ml) was shaken on a water bath at 37 °C for 30 d. The mixture was evaporated *in vacuo* to dryness and the residue was chromatographed on silica gel [hexane–EtOAc (3 : 2) and $CHCl_3$–MeOH–H_2O (7 : 3 : 0.5)] to give **6a** (10 mg) and the sugar fraction. From the sugar fraction, the presence of D-glucose was demonstrated as for **1**.

2α,7,8β-Trihydroxykessane (6a) Colorless needles (aq. MeOH), mp 213—215 °C, $[\alpha]_D^{21}$ $+10.0°$ (c=0.5, MeOH). Positive FAB-MS *m/z*: 271.1901 $[M+H]^+$ (Calcd for $C_{15}H_{27}O_4$: 271.1909), 253 $[M-H_2O+H]^+$, 235 $[M-2H_2O+H]^+$ (base), 217 $[M-3H_2O+H]^+$. Negative FAB-MS *m/z*: 269 $[M-H]^-$ (base).

Acetylation of 6a Aglycone **6a** (5 mg) was acetylated with Ac_2O and pyridine at room temperature for 12 h to give a diacetate of **6a** (**6a′**: 6 mg) as an amorphous powder. ^{1}H-NMR ($CDCl_3$) δ: 0.93 (3H, d, J=7.5 Hz, H_3-15), 1.05 (1H, dddd, J=14.5, 5.5, 2.5 Hz, H-3α), 1.16 (3H, s, H_3-14), 1.26 (3H, s, H_3-13), 1.31 (3H, s, H_3-12), 1.71 (1H, br dd, J=13.5, 13.5 Hz, H-6α), 1.98 (1H, dd, J=14.5, 8.0 Hz, H-9β), 2,04 (1H, dd, J=13.5, 6.5 Hz, H-6β), 2.05 (1H, dd, J=13.5, 9.5 Hz, H-1), 2.06 (1H, m, H-4), 2.24 (1H, br ddd, J=13.5, 13.5, 6.5 Hz, H-5), 2.46 (1H, dd, J=14.5, 9.5 Hz, H-9α), 2.61 (1H, ddd, J=14.5, 9.0, 9.0 Hz, H-3β), 4.92 (1H, ddd, J=9.5, 9.0, 5.5 Hz, H-2), 4.98 (1H, dd, J=9.5, 8.0 Hz, H-8), 1.99, 2.13 (each 3H, s, OAc). ^{13}C-NMR ($CDCl_3$) δ: 52.69 (C-1), 75.31 (C-2), 39.80 (C-3), 31.58 (C-4), 37.55 (C-5), 39.46 (C-6), 71.51 (C-7), 74.88 (C-8), 44.01 (C-9), 71.81 (C-10), 77.81 (C-11), 26.79 (C-12), 24.86 (C-13), 27.37 (C-14), 17.88 (C-15), acetoxyl [21.21, 21.28, 170.37, 170.50].

2α,7-Dihydroxykessane 2-*O*-β-D-Glucopyranoside (7) An amorphous powder, $[\alpha]_D^{21}$ $+17.6°$ (c=1.4, MeOH). Positive FAB-MS *m/z*: 834 $2[M+H]^+$, 417.2487 $[M+H]^+$ (Calcd for $C_{21}H_{37}O_8$: 417.2489), 399 $[M-H_2O+H]^+$ (base), 237 $[M-C_6H_{12}O_6+H]^+$. Negative FAB-MS *m/z*: 415 $[M-H]^-$ (base).

Enzymatic Hydrolysis of 7 A mixture of **7** (50 mg) and hesperidinase (5 mg) in water (5 ml) was treated as described for **6**, and the residue was chromatographed on silica gel [hexane–EtOAc (3 : 2) and $CHCl_3$–MeOH–H_2O (7 : 3 : 0.5)] to **7a** (16 mg) and the sugar fraction. From the sugar fraction, the presence of D-glucose was demonstrated as for **1**.

2α,7-Dihydroxykessane (7a) An amorphous powder, $[\alpha]_D^{21}$ $+11.6°$ (c=0.4, MeOH). Positive FAB-MS *m/z*: 277 $[M+Na]^+$, 255.1968 $[M+H]^+$ (Calcd for $C_{15}H_{27}O_3$: 255.1960), 235 $[M-H_2O+H]^+$ (base). Negative FAB-MS *m/z*: 253 $[M-H]^-$ (base).

2α,7,12-Trihydroxykessane 2-*O*-β-D-Glucopyranoside (8) An amorphous powder, $[\alpha]_D^{23}$ $+10.0°$ (c=0.8, MeOH). Positive FAB-MS *m/z*: 455.2252 $[M+Na]^+$ (base, Calcd for $C_{21}H_{36}O_9Na$: 455.2257), 253 $[M-C_6H_{12}O_6+H]^+$. Negative FAB-MS *m/z*: 431 $[M-H]^-$ (base).

(1*R*,4*R*,5*R*,7*R*,10*S*)-10,11,15-Trihydroxyguaiane 11-*O*-β-D-Glucopyranoside (9) An amorphous powder, $[\alpha]_D^{21}$ $+9.8°$ (c=1.3, MeOH). Positive FAB-MS *m/z*: 838 $2[M+H]^+$, 441.2437 $[M+Na]^+$ (Calcd for $C_{21}H_{38}O_8Na$: 441.2465), 419 $[M+H]^+$, 401 $[M-H_2O+H]^+$ (base), 239 $[M-C_6H_{12}O_6+H]^+$. Negative FAB-MS *m/z*: 417 $[M-H]^-$ (base).

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