[Chem. Pharm. Bull.
36(8)2990—2992(1988)]

Minor Constituents from the Seeds of Japanese Star-Anise

ISAO KOUNO,* TOSHIHIKO AKIYAMA, and NOBUSUKE KAWANO

*Faculty of Pharmaceutical Sciences, Nagasaki University,
1-14 Bunkyo-machi, Nagasaki 852, Japan*

(Received February 22, 1988)

Two sesquiterpene lactones, 6-deoxypseudoanisatin and a new compound have been isolated from the seeds of Japanese star-anise along with three known compounds, anisatin, neoanisatin, and pseudoanisatin. The new compound was named 6-deoxymajucin and its structure was deduced by spectral studies.

Keywords——*Illicium anisatum*; Illiciaceae; sesquiterpene; pseudoanisatin; anisatin; 6-deoxypseudoanisatin; 6-deoxymajucin

Three sesquiterpene lactones, anisatin (**1**), neoanisatin (**2**),[1,2] and pseudoanisatin (**3**),[3] have been isolated from Japanese star-anise, the fruits of *Illicium anisatum* L. (Illiciaceae), a well known toxic plant in Japan. We now report two additional compounds obtained from the seeds of this plant: 6-deoxypseudoanisatin (**4**) and a new compound named 6-deoxymajucin (**5**). These two compounds, together with **1**, **2**, and **3**, were isolated from the ethanol extracts of the seeds by counter-current distribution and chromatographic separations.

6-Deoxypseudoanisatin (**4**) was identified by direct comparison with the recently isolated[4] compound from the bark of *I. dunnianum*. The new compound (**5**) was obtained as colorless needles, mp 267—270 °C. In its mass spectrum (MS), the molecular ion (m/z 312) indicated a molecular formula of $C_{15}H_{20}O_7$, one oxygen atom less than that of anisatin (**1**). The infrared spectrum (IR) showed the presence of γ-lactone (1740 cm^{-1}) and δ-lactone (1715 cm^{-1}) moieties in the new compound.

The proton nuclear magnetic resonance (^{1}H-NMR) spectrum of **5** (Table I) showed two

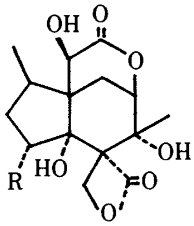
TABLE I. ^{1}H-NMR Chemical Shifts for Compounds **5** and **6** in C_5D_5N[a)]

	5	**6**
H-1	2.90 (ddq, J=10.3, 9.2, 7.3)	3.02 (ddq, J=10.2, 9.5, 7.0)
H-2α	2.15 (ddd, J=12.5, 10.3, 4.0)	2.21 (ddd, J=12.6, 10.2, 4.4)
H-2β	2.40 (dt, J=12.5, 9.2)	2.48 (dt, J=12.6, 9.5)
H-3	5.14 (dd, J=9.2, 4.0)	5.21 (dd, J=9.5, 4.4)
H-6	2.93 (d, J=5.5)	—
H-7	5.18 (ddd, J=5.5, 3.7, 2.2)	5.14 (dd, J=3.3, 2.2)
H-8α	2.63 (dd, J=13.9, 2.2)	3.11 (dd, J=14.3, 2.2)
H-8β	2.01 (dd, J=13.9, 3.7)	2.05 (dd, J=14.3, 3.3)
H-10	4.56 (br d, J=5.1)	4.65 (br d, J=4.5)
H-13	1.66 (br s)	1.95 (br s)
H-14a	4.18 (d, J=10.6)	4.30 (d, J=10.8)
H-14b	5.06 (br d, J=10.6)	5.11 (br d, J=10.8)
H-15	1.07 (d, J=7.3)	1.10 (d, J=7.0)

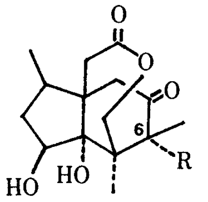
a) Run at 400 MHz. Chemical shifts are given on the δ (ppm) scale, and coupling constants are given in Hz (s, singlet; d, doublet; t, triplet; q, quartet; br, broad).

NII-Electronic Library Service

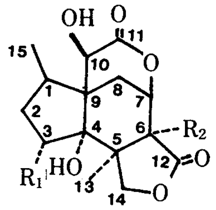
methyl signals (δ 1.07 and 1.66), three pairs of methylene signals (δ 2.40 and 2.15; δ 2.01 and 2.63; δ 4.18 and 5.06) and three proton signals (δ 4.56, 5.14, and 5.18). The two-dimensional proton–proton correlation (2D ^{1}H–^{1}H COSY) spectrum of **5** indicated the following proton connectivities: the methyl signal (δ 1.07, 3H, d, $J=7.3$ Hz)–the methine signal (δ 2.90, 1H, ddq, $J=10.3$, 9.2, 7.3 Hz)–the methylene signals (δ 2.40 1H, dt, $J=12.5$, 9.2 Hz, and δ 2.15, 1H, ddd, $J=12.5$, 10.3, 4.0 Hz)–the methine signal (δ 5.14, 1H, dd, $J=9.2$, 4.0 Hz). The methyl signal at δ 1.66 (3H, br s) was weakly coupled with one of the methylene signals of the γ-lactone (δ 5.06 1H, br d, $J=10.6$ Hz). These spectral data suggested a closely related structure to majucin (**6**), which was recently isolated from *I. majus* together with the 3-deoxy compound, neomajucin (**7**). The structure of **7** has been established[5] by an X-ray crystallographic analysis.



1: R=OH
anisatin
2: R=H
neoanisatin



3: R=OH
pseudoanisatin
4: R=H
6-deoxypseudoanisatin



5: R_1=OH, R_2=H
6-deoxymajucin
6: $R_1=R_2$=OH
majucin
7: R_1=H, R_2=OH
neomajucin

TABLE II. ^{13}C-NMR Data for Compounds **5** and **6** in C_5D_5N (100 MHz; δ ppm)

	5	6		5	6
C-1	38.1 (d)	38.0 (d)	C-9	51.5 (s)	51.5 (s)
C-2	42.7 (t)	42.9 (t)	C-10	70.2 (d)	70.3 (d)
C-3	72.4 (d)	72.7 (d)	C-11	175.8 (s)	174.7 (s)
C-4	82.1 (s)	82.8 (s)	C-12	175.3 (s)	177.6 (s)
C-5	45.1 (s)	47.5 (s)	C-13	25.5 (q)	20.9 (q)
C-6	54.1 (d)	79.9 (s)	C-14	74.3 (t)	72.4 (t)
C-7	75.3 (d)	80.6 (d)	C-15	14.2 (q)	14.1 (q)
C-8	29.5 (t)	27.1 (t)			

Assignments were made on the basis of 2D ^{1}H–^{13}C COSY spectra and 2D long-range ^{1}H–^{13}C COSY spectra.

In the 2D ^{1}H–^{1}H COSY spectrum of **5**, a signal at δ 5.18 (1H, ddd, $J=5.5$, 3.7, 2.2 Hz) was coupled with the signals of δ 2.01 (H-8β), 2.63 (H-8α), and 2.93 (H-6), and should be assigned to H-7, indicating a 6-deoxy structure for the new compound. An upfield-shifted proton signal assigned to H-8α (δ 3.11 to δ 2.63) also supports the 6-deoxy structure of **5**. In the carbon-13 nuclear magnetic resonance (^{13}C-NMR) spectrum of **5** (Table II), the signal of C-6 was seen as a doublet at δ 54.1 as compared with a singlet (δ 79.9) in the case of majucin (**6**). Therefore, the structure of the new compound was deduced to be 6-deoxymajucin.

This is the first report on the isolation of 6-deoxypseudoanisatin (**4**) and 6-deoxymajucin (**5**) from the seeds of Japanese star-anise. It is noteworthy that the seeds of Japanese star-anise contain all kinds of *Illicium* sesquiterpene lactones, *i.e.* anisatin, pseudoanisatin, and majucin types.

NII-Electronic Library Service

Experimental

The melting point was determined on a micro-hot stage (Yanagimoto) and is uncorrected. The IR spectrum was recorded on a Shimadzu IR-408 spectrometer, and ^{1}H–, ^{13}C-NMR, and 2D ^{1}H–^{1}H COSY spectra on a JEOL GX-400 spectrometer (with tetramethylsilane as an internal standard). The MS was taken with a JEOL DX-303 spectrometer. Thin layer chromatography (TLC) was performed on precoated Silica gel $60F_{254}$ plates (Merck), and column chromatography on Silica gel type 60 (Merck).

Isolation of the Constituent——The powdered seeds of *Illicium anisatum* (4.2 kg) were extracted with MeOH at room temperature to give the MeOH extract, which was suspended in water and defatted with *n*-hexane. The water layer was extracted with AcOEt to afford the AcOEt-soluble part (67 g), which was subjected to counter-current distribution using the solvent system of AcOEt–H_2O. Of the combined fractions I—IV, fraction II was subjected to chromatography on a silica gel column ($CHCl_3$: MeOH = 97 : 3) to give pseudoanisatin (**3**) (2.2 g) and neoanisatin (**2**) (10 mg). Fraction III was chromatographed on a silica gel column ($CHCl_3$: MeOH = 24 : 1 then $CHCl_3$: AcOEt = 1 : 1) to give anisatin (**1**) (550 mg), 6-deoxymajucin (**5**) (11 mg), and 6-deoxypseudoanisatin (**4**) (15 mg).

Spectral data for anisatin (**1**), neoanisatin (**2**), pseudoanisatin (**3**), and 6-deoxypseudoanisatin (**4**) were identical with those of authentic samples.

6-Deoxymajucin (**5**): Colorless needles. mp 267—270 °C (AcOEt). MS *m/z*: 312 (M^+). IR $\nu_{\max}^{\text{Nujol}}$ cm^{-1}: 3525, 1740, 1715. $[\alpha]_D^{19}$ $-40.0°$ ($c = 0.13$, dioxane). ^{1}H- and ^{13}C-NMR data are given in Tables I and II, respectively.

Acknowledgments We wish to thank Prof. K. Yamada of Nagoya University for the authentic sample of neoanisatin. We are also indebted to Mr. Y. Ohama for the measurement of 400 MHz NMR spectra.

References

1) K. Yamada, S. Takada, S. Nakamura, and Y. Hirata, *Tetrahedron*, **24**, 199; 1267 (1968).
2) K. Yamada, S. Takada, and Y. Hirata, *Tetrahedron*, **24**, 1255 (1968).
3) I. Kouno, H. Irie, and N. Kawano, *J. Chem. Soc., Perkin Trans 1*, **1984**, 2511.
4) I. Kouno, N. Kawano, and C.-S. Yang, *J. Chem. Soc., Perkin Trans 1*, **1988**, 1537.
5) C.-S. Yang, I. Kouno, and N. Kawano, *Tetrahedron Lett.*, **29**, 1165 (1988).