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The Constituents of *Schizandra chinensis* BAILL. VIII.¹⁾ The Structures of Two New Lignans, Tigloylgomisin P²⁾ and Angeloylgomisin P

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Two new dibenzocyclooctadiene lignans, tigloylgomisin P(1), and angeloylgomisin P(2) were isolated from the fruits of *Schizandra chinensis* BAILL. (Schizandraceae). Their absolute structures were elucidated by means of chemical and spectral studies.

Keywords—*Schizandra chinensis* BAILL.; Schizandraceae; dibenzocyclooctadiene; lignan; tigloylgomisin P; angeloylgomisin P; NOE; ¹³C-NMR

In the previous papers of this series, we have reported a number of dibenzocyclooctadiene lignans isolated from the fruits of *Schizandra chinensis* BAILL. (Schizandraceae).⁴⁾ This paper deals with two additional new lignans, tigloylgomisin P (1, yield 0.0013%) and angeloylgomisin P (2, 0.0008%) from the same source.

Tigloylgomisin P (1) was isolated as a white amorphous powder, C₂₈H₃₄O₉, [α]_D²⁵ -64.2° (CHCl₃). The ultraviolet (UV) spectrum of 1, with absorption maxima at 217 (log ϵ 4.68), 255 (sh 3.99) and 282 nm (3.51), and the infrared (IR) spectrum, with bands at 3480 (OH), 1712 (ester), 1645 (C=C), 1616 and 1595 (aromatic) cm⁻¹, indicate that 1 is a dibenzocyclooctadiene lignan⁴⁾ possessing a hydroxyl and an ester linkage. The proton nuclear magnetic resonance (¹H-NMR) spectrum (Table I) and the carbon (¹³C)-NMR spectrum (Table II) indicate the presence of a methylenedioxy moiety and four methoxyl groups on the aromatic rings and also the presence of a tertiary methyl group attached to a carbon carrying a hydroxyl group (CH₃-C-OH), a secondary methyl group, a benzylic methine and a tigloyl group [¹³C-NMR, δ in CDCl₃, tigloyl: 12.2 (α -Me), 14.4 (β -Me), 128.8, 137.5 (>C=C<), 166.6 (>C=O)].⁵⁾ The mass spectral analysis also supports the presence of a tigloyl group in 1 (see "Experimental").

On hydrolysis with 3% ethanolic potassium hydroxide, 1 afforded a tiglic acid and a diol (3), named gomisin P, C₂₃H₂₈O₈, mp 114–116°, [α]_D²⁵ -94.3° (CHCl₃). The circular dichroism (CD) spectrum of 3 shows a positive Cotton effect at 221 nm ([θ] +62000) and two negative Cotton effects at 242 nm ([θ] -66000, sh) and 253 nm ([θ] -76000), indicating that 3 possesses an S-biphenyl configuration.^{4a)}

The singlet at δ 4.32 in the ¹H-NMR spectrum of 3, which appeared at δ 5.37 (in C₆D₆) (δ 5.54 in CDCl₃) in 1, was assigned to a benzylic methine. This shows that the tigloyl group in 1 is linked to a benzylic hydroxyl group. On the basis of a comparison of the ¹H-NMR spectrum of 3 with that of deangeloylgomisin B (4),^{4a)} 3 is assumed to be a diastereomer of 4 at the C-6 and C-7 positions, as discussed below. In the ¹H-NMR spectrum, the tertiary

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Chart 1

TABLE I. ^1H -NMR Spectral Data [δ in CDCl_3 , 60 MHz for 2,3,6 and 100 MHz for 1,4]

Compd.	4-H, s 11-H, s	6-H s	OCH_2O s	OCH_3 s	$9\alpha\text{-H}$ dd ($J = \text{Hz}$)	$9\beta\text{-H}$ dd ($J = \text{Hz}$)	6-OH ^{c)} s	$\text{HO}^c\text{-C}_{(7)}\text{-Me}$ s s	$\text{H-C}_{(8)}\text{-Me}^d$ m d ($J = \text{Hz}$)
1 ^{a, b)}	6.95 6.13	5.90	5.37	3.53, 3.70 3.83, 3.92	2.10 (center) (2H, m)	—	—	2.07 1.13	1.98 1.10 (7)
2 ^{a, b)}	6.97 6.49	5.97	5.37	3.53, 3.71 3.85, 3.92	2.10 (center) (2H, m)	—	—	1.80 1.11	1.88 1.08 (7)
3	7.10 6.47	4.32	5.93	3.58, 3.83 3.92 ($\times 2$)	2.10 (center) (2H, m)	—	2.40	2.40 1.00	1.77 1.07 (7)
4	6.62 6.51	4.59	5.95	3.61, 3.90 3.93 ($\times 2$)	2.35 (12/8.5)	2.05 (12/1)	1.56	1.56 1.40	1.85 1.11 (7)
6 ^{b)}	7.07 6.50	4.57	5.99	3.62, 3.78 3.93, 3.95	2.10 (center) (2H, m)	—	—	0.83	1.77 1.08 (6)

a) Measured in C_6D_6 .b) Other signals: 1: $\begin{array}{c} \text{Me} \quad \beta \quad \alpha \quad \text{Me} \\ \diagdown \quad \text{C}=\text{C} \quad \diagup \\ \text{H} \quad \diagdown \quad \text{CO} \end{array}$: 1.38 (3H, dq, $J = 7/1$ Hz, $\beta\text{-Me}$), 1.68 (3H, m, $\alpha\text{-Me}$), 6.85 (1H, m, $\beta\text{-H}$).2: $\begin{array}{c} \text{Me} \quad \beta \quad \alpha \quad \text{CO} \\ \diagdown \quad \text{C}=\text{C} \quad \diagup \\ \text{H} \quad \diagdown \quad \text{Me} \end{array}$: 1.80 (3H, s, $\alpha\text{-Me}$), 1.87 (3H, dq, $J = 7/1.5$ Hz, $\beta\text{-Me}$), 5.70 (1H, m, $\beta\text{-H}$).3: $\begin{array}{c} \text{O} \quad \diagdown \quad \text{Me} \\ \diagdown \quad \text{C} \quad \diagup \\ \text{O} \quad \diagdown \quad \text{Me} \end{array}$: 1.27, 1.55 (each 3H, s)c) Confirmed by addition of D_2O .

d) Confirmed by decoupling experiments.

e) d=doublet, m=multiplet, q=quartet, s=singlet.

methyl of 3 appears at higher field than that of 4 (δ 1.40), indicating that it is shielded by the aromatic rings, and one of the aromatic protons (δ 7.10) appears at lower field than that of 4 (δ 6.62), suggesting the presence of a C-6 α -oriented hydroxyl group. This view is also supported by the ^{13}C -NMR spectra of 3 and 4.^{4f)}

Oxidation of 3 with chromium trioxide in pyridine afforded a ketoaldehyde (5),^{4a)} $\text{C}_{23}\text{H}_{26}\text{O}_8$, which was identical with 5 prepared from 4 by oxidation with chromium trioxide in acetic acid. This further supports the above considerations. On the other hand, treatment of 3 with CuSO_4 and 0.25% H_2SO_4 (in acetone) afforded an acetonide (6), $\text{C}_{26}\text{H}_{32}\text{O}_8$ (M^+ , m/z 472), $[\alpha]_D^{25} -135^\circ$ (CHCl_3). Dreiding model examination indicated that the structure of the acetonide is represented by the formula 6.

Finally, the structure of 1, including the conformation of the cyclooctadiene ring, was confirmed by measurements of intramolecular nuclear Overhauser effects (NOE) (in C_6D_6), as shown in Fig. 1.^{4a, b)} Irradiation of a methoxyl signal (δ 3.53) caused a 14% increase in the integrated intensity of the lower field aromatic proton signal at δ 6.95 ($\text{C}_{(4)}\text{-H}$), while the higher field aromatic proton at δ 6.13 ($\text{C}_{(11)}\text{-H}$) was unaffected by irradiation of any methoxyl signal. Irradiation of the benzylic methylene protons (irradiated at δ 2.01) caused a 13% increase in the integrated intensity of the $\text{C}_{(11)}$ -proton signal, while the $\text{C}_{(4)}$ -proton was unaf-

TABLE II. ^{13}C -NMR Spectral Data for 1 and 2 [δ in CDCl_3 , ^{13}C : 20 MHz at 25°]

Carbon	1	2	Carbon	1	2
1	151.1 (s) ^a	151.1 (s) ^a	17	18.8 (q)	18.8 (q)
2	141.1 (s) ^b	141.1 (s) ^b	18	17.5 (q)	17.5 (q)
3	152.4 (s) ^a	152.4 (s) ^a	C-1, 14	60.6, 59.9 (q)	60.6, 59.9 (q)
4	106.2 (d)	106.5 (d)	C-2	60.9 (q)	60.9 (q)
5	133.1 (s)	132.9 (s)	C-3	55.9 (q)	55.9 (q)
6	77.6 (d)	77.3 (d)	OCH_2O	101.0 (t)	101.0 (t)
7	75.2 (s)	75.2 (s)	α -Me	12.2 (q)	
8	46.6 (d)	46.7 (d)	β -Me	14.4 (q)	
9	36.7 (t)	36.7 (t)	C=C	128.8 (s)	
10	136.7 (s)	136.7 (s)		137.5 (d)	
11	103.0 (d)	103.0 (d)	C=O	166.6 (s)	
12	149.5 (s)	149.5 (s)	α -Me		15.8 (q)
13	135.6 (s)	135.7 (s)	β -Me		20.8 (q)
14	141.5 (s) ^b	141.5 (s) ^b	C=C		127.6 (s)
15	122.9 (s) ^c	122.9 (s) ^c			138.7 (d)
16	119.6 (s) ^c	119.6 (s) ^c	C=O		166.4 (s)

a, b, c) Assignments within any vertical column may be reversed.

d) Tiglic acid (δ in CDCl_3): 14.5 11.6
 $\text{Me} \searrow \text{C} = \text{C} \nearrow \text{Me}$
 $\text{H} \backslash 140.0 \quad 128.3 \text{ COOH}$
 $\text{H} \backslash 174.0$

e) Angelic acid (δ in CDCl_3): 20.3 170.1
 $\text{Me} \searrow \text{C} = \text{C} \nearrow \text{COOH}$
 $\text{H} \backslash 141.1 \quad 127.3 \text{ Me}$
 $\text{H} \backslash 16.0$

f) d=doublet, q=quartet, s=singlet, t=triplet.

ected by irradiation of the benzylic methine (δ 5.90, $\text{C}_{(6\beta)}\text{-H}$). Irradiation of the $\text{C}_{(8)}$ -methine (δ 1.98) caused a 13% increase in the integrated intensity of the $\text{C}_{(6)}$ -methine signal (δ 5.90). These findings indicate that the methylenedioxy moiety is located adjacent to the $\text{C}_{(11)}$ -proton, and that the $\text{C}_{(6)}$ - and $\text{C}_{(8)}$ -protons are β -oriented. Irradiation of the tertiary methyl signal (δ 1.13) affected the $\text{C}_{(4)}$ -proton, indicating that the tertiary methyl and the $\text{C}_{(4)}$ -proton must be close to each other. On the other hand, the aromatic protons were unaffected by irradiation of the secondary methyl signal (δ 1.10).

Fig. 1. NOE in 1 (in C_6D_6)

On the basis of the above results, the absolute structure of tigloylgomisin P was elucidated as 1, possessing a twist-boat-chair conformation of the cyclooctadiene ring.

Angeloylgomisin P (2) was isolated as a white amorphous powder, $\text{C}_{28}\text{H}_{34}\text{O}_9$, $[\alpha]_{\text{D}}^{25} -98.5^\circ$ (CHCl_3). The UV, mass, ^1H -NMR and ^{13}C -NMR spectra of 2 indicate that 2 is a dibenzocyclooctadiene lignan possessing the same skeleton as 1 and an angeloyl group [^{13}C -NMR, δ in CDCl_3 , angeloyl: 15.8 (α -Me), 20.8 (β -Me), 127.6, 138.7 (C=C), 166.4 (C=O)].⁵ On hydrolysis with 3% ethanolic potassium hydroxide, 2 gave 3 and angelic acid.⁶ The structure of angeloylgomisin P was thus elucidated as 2.

6) Angelic acid was partly isomerized to tiglic acid during hydrolysis.

Experimental

All melting points were determined on a Yanagimoto micromelting point apparatus (a hot stage type) and are uncorrected. The UV spectra were recorded with a Hitachi 624 digital spectrophotometer and the IR spectra with a Hitachi EPI-G2 machine. The ^1H -NMR spectra were recorded with Varian T-60 and JEOL PS-100 spectrometers and ^{13}C -NMR spectra with a Varian FT-80A spectrometer, with tetramethylsilane as an internal standard. The mass spectra were measured with a Hitachi double-focusing mass spectrometer. The specific rotations were measured with a JASCO DIP-SL unit and the circular dichroism (CD) spectrum with a JASCO J-20. Gas liquid chromatography (GLC) was carried out on a Hitachi 073 gas chromatograph with FID. For silica gel column chromatography, Kieselgel 60 (Merck) was used. Thin-layer chromatography (TLC) was carried out on Merck plates precoated with Kieselgel 60F₂₅₄. Preparative layer chromatography (PLC) was carried out on plates (20 × 20 cm) coated with Kieselgel PF₂₅₄ (Merck).

Isolation of 1 and 2—In the previous paper,^{4a)} it was reported that the pet. ether and methanolic extracts of the fruits of *Schizandra chinensis* BAILL. (4.671 kg) afforded twelve fractions (fr. 1–12) on silica gel column chromatography, developing with hexane, acetone–benzene and acetone–solvent systems. Fr. 6 (10.97 g) was rechromatographed on silica gel (240 g, 4.5 × 29 cm) with a hexane–EtOAc solvent system. The fractions eluted with hexane–EtOAc (4:1) were concentrated to give a residue (1104 mg), which was rechromatographed on silica gel (30 g, 2 × 17 cm) with a benzene–ether solvent system. The fractions eluted with benzene–ether [(49:1) and (24:1)] were combined and concentrated to give a residue (385 mg). Repeated PLC [i) hexane–acetone (7:3), *Rf* 0.62, ii) benzene–ether (2:1), *Rf* 0.67, iii) CHCl_3 –EtOH (20:1), *Rf* 0.91] of this residue gave 2 (39 mg, yield 0.0008%). The fractions eluted with benzene–ether (47:3) were concentrated to give a residue (548 mg), which was purified by PLC [hexane–ether (1:2), *Rf* 0.47] to give 1 (24 mg). The fractions eluted with hexane–EtOAc (3:1) were concentrated to give a residue (238 mg). Repeated PLC [i) benzene–ether (7:3), *Rf* 0.56; ii) hexane–ether (1:2), *Rf* 0.47] of this residue gave 1 (35 mg, 0.0013%).

Tigloylgomisin P(1)—Pure tigloylgomisin P was obtained as a white amorphous powder (from ether–hexane), $[\alpha]_D^{25} -64.2^\circ$ ($c=0.436$, CHCl_3). UV $\lambda_{\text{max}}^{\text{EtOH}}$ nm (log ϵ): 217 (4.68), 255 (sh 3.99), 282 (3.51). IR $\nu_{\text{max}}^{\text{KBr}}$ cm^{-1} : 3480 (OH), 1712 (ester), 1645 (C=C), 1616, 1595 (aromatic). High resolution MS (m/z), Calcd for $\text{C}_{28}\text{H}_{34}\text{O}_9(\text{M}^+)$: 514.2208. Found: 514.2180. MS, m/z (%): 514 (M^+ , 12), 414 [$\text{M}^+ - \text{CH}_3\text{CH}=\text{C}(\text{CH}_3)\text{COOH}$, 19], 343 (43), 83 [$\text{CH}_3\text{CH}=\text{C}(\text{CH}_3)\text{CO}^+$, 100], 55 ($\text{CH}_3\text{CH}=\text{C}^+ - \text{CH}_3$, 90). Anal. Calcd for $\text{C}_{28}\text{H}_{34}\text{O}_9$: C, 65.36; H, 6.66. Found: C, 65.62; H, 6.85. The ^1H -NMR and ^{13}C -NMR data are given in Tables I and II, respectively.

Angeloylgomisin P(2)—Pure angeloylgomisin P was obtained as a white amorphous powder, $[\alpha]_D^{25} -98.5^\circ$ ($c=2.18$, CHCl_3). UV $\lambda_{\text{max}}^{\text{EtOH}}$ nm (log ϵ): 3500 (OH), 1720 (ester), 1644 (C=C), 1618, 1598 (aromatic). High resolution MS (m/z), Calcd for $\text{C}_{28}\text{H}_{34}\text{O}_9(\text{M}^+)$: 514.2208. Found: 514.2202. MS, m/z (%): 514 (M^+ , 28), 414 [$\text{M}^+ - \text{CH}_3\text{CH}=\text{C}(\text{CH}_3)\text{COOH}$, 36], 343 (57), 83 [$\text{CH}_3\text{CH}=\text{C}(\text{CH}_3)\text{CO}^+$, 73], 55 ($\text{CH}_3\text{CH}=\text{C}^+ - \text{CH}_3$, 100). The ^1H -NMR and ^{13}C -NMR data are given in Tables I and II, respectively.

Hydrolysis of 1—A solution of 1 (38 mg) in 3% KOH–EtOH (2 ml) was kept at 55° for 2 hr, then diluted with H_2O (20 ml) and extracted with ether (15 ml × 3). The combined ethereal extract was washed with H_2O , dried over Na_2SO_4 and concentrated to give a residue, which was purified by PLC [benzene–ether (3:2)] to give a diol (3), colorless needles (from ether–hexane), mp 114–116°, $[\alpha]_D^{25} -94.3^\circ$ ($c=0.530$, CHCl_3) (30 mg). UV $\lambda_{\text{max}}^{\text{EtOH}}$ nm (log ϵ): 220 (4.59), 254 (sh 4.02), 283 (sh 3.49), 294 (sh 3.37). IR $\nu_{\text{max}}^{\text{KBr}}$ cm^{-1} : 3440 (OH), 1615, 1595 (aromatic). CD ($c=0.0183$, MeOH), $[\theta]^{25}$ (nm): +62000 (221), –66000 (sh 242), –76000 (253). Anal. Calcd for $\text{C}_{28}\text{H}_{28}\text{O}_8$: C, 63.88; H, 6.53. Found: C, 63.80; H, 6.63.

The aqueous solution was acidified with 1 N HCl and extracted with ether. The ethereal extract was washed with H_2O , dried over Na_2SO_4 and concentrated to give a residue, which was sublimed (70°, 15 mmHg) to give colorless needles (2 mg), mp 63–64°. This compound was identified as tiglic acid by direct comparison with an authentic sample [GLC, IR and mixed mp; GLC conditions: column, SP-1200 (10%) + H_3PO_4 (1%) on Chromosorb WAW (80–100 mesh) 3 mm × 2 m; column temperature, 130°; injection temperature, 150°; carrier gas, N_2 , 30 ml/min; tiglic acid, t_R (min), 8.7].

Oxidation of 3 with Chromium Trioxide in Pyridine— CrO_3 (25 mg) was added to a solution of 3 (12 mg) in pyridine (0.5 ml). The reaction mixture was stirred at 15° for 30 min, then diluted with H_2O and extracted with ether (15 ml × 3). The combined ethereal extract was washed with 1 N HCl, then H_2O , dried over Na_2SO_4 and concentrated. The residue was purified by PLC [hexane–acetone (7:3)] to give 5 as a colorless oil (8 mg), $[\alpha]_D^{25} \approx 0^\circ$ ($c=0.277$, CHCl_3). The ^1H -NMR of 5 was identical with that of an authentic sample.^{4a)} Treatment of 5 (5 mg) with 1% 2,4-dinitrophenylhydrazine in 2 N HCl–EtOH (2:1) gave reddish needles (from acetone–EtOH), mp 129.5–132.5° (6 mg). This compound was identical with the di-2,4-dinitrophenylhydrazone of 5 on direct comparison (^1H -NMR and mixed mp).

Preparation of 6—Anhydrous CuSO_4 (80 mg) and 0.25% H_2SO_4 (in acetone) (0.05 ml) were added to a solution of 3 (11 mg) in dry acetone (4 ml). The reaction mixture was stirred at 22° for 3 hr, then diluted with H_2O (20 ml) and extracted with ether (15 ml × 3). The combined ethereal extract was washed with H_2O , dried over Na_2SO_4 and concentrated to dryness. The residue was purified by PLC [benzene–ether

(4:1)] to give **6** as a white amorphous powder, $[\alpha]_D^{25} -135^\circ$ ($c=0.615$, CHCl_3). IR $\nu_{\text{max}}^{\text{KBr}}$ cm^{-1} : 1619, 1599 (aromatic). MS, m/z (%): 472 [$\text{C}_{26}\text{H}_{32}\text{O}_8(\text{M}^+)$, 93], 457 (M^+-CH_3 , 100), 414 (10), 343 (43).

Hydrolysis of 2—A solution of **2** (27 mg) in 3% KOH-EtOH (1 ml) was kept at 50° for 1.5 hr, then the reaction mixture was treated as described for the hydrolysis of **1** to give a diol and an acid. The former was obtained as colorless needles (17 mg), mp $113.5\text{--}116^\circ$, $[\alpha]_D^{25} -96.7^\circ$ ($c=1.62$, CHCl_3). Anal. Calcd for $\text{C}_{23}\text{H}_{28}\text{O}_8$: C, 63.88; H, 6.53. Found: C, 64.10; H, 6.60. This compound was identified as **3** by direct comparison with an authentic sample (IR, $[\alpha]_D$ and mixed mp). The latter was sublimed (70° , 15 mmHg) to give a colorless oil (1 mg). The presence of angelic acid and tiglic acid in this sublimate in a ratio of 3:1 was demonstrated by GLC.⁶⁾ GLC conditions were the same as in the case of **1**; angelic acid, $t_R(\text{min})$, 6.5; tiglic acid, $t_R(\text{min})$, 8.4.

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