Phytochemistry, Vol. 27, No. 2, pp. 569–573, 1988.
Printed in Great Britain.

0031–9422/88 $3.00+0.00

A LIGNAN FROM *SCHIZANDRA CHINENSIS**

YUKINOBU IKEYA, HEIHACHIRO TAGUCHI, HIROSHI MITSUHASHI, SHIGEFUMI TAKEDA†, YOSHIO KASE† and MASAKI ABURADA†

Tsumura Laboratory, 3586 Yoshiwara Ami-Machi, Inashiki-Gun, Ibaraki, Japan; †Tsumura Research Institute for Pharmacology, 3586 Yoshiwara Ami-Machi, Inashiki-Gun, Ibaraki, Japan

(*Revised received* 17 *June* 1987)

Key Word Index—*Schizandra chinensis*.; Schizandraceae; dibenzocyclooctadiene; lignan; isoschizandrin; anti-ulcer effect.

Abstract—A new dibenzocyclooctadiene lignan, isoschizandrin was isolated from the fruits of *Schizandra chinensis*. The structure was elucidated on the basis of the spectral analysis and chemical correlation with schizandrin and (+)-deoxyschizandrin. In experiments using rats, schizandrin and its derivatives, including isoschizandrin, showed inhibitory effects on stress-induced gastric ulceration (p.o.).

INTRODUCTION

Thirty-four dibenzocyclooctadiene lignans[1] have been isolated from the fruits of *Schizandra chinensis* Baill. This paper deals with the isolation and structure elucidation of the new lignan, isoschizandrin (**1a**) and its inhibitory effect on stress-induced gastric ulceration.

RESULTS AND DISCUSSION

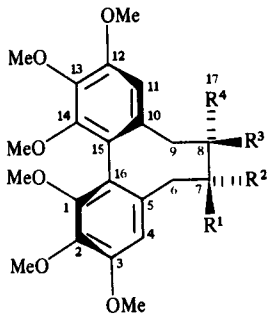
Isoschizandrin (**1a**) was obtained as a white amorphous powder, $C_{24}H_{32}O_7$, $[\alpha]_D+92°$ ($CHCl_3$). The UV, IR, ^1H NMR (Table 1) and ^{13}C NMR (Table 2) spectra of **1a** indicated that **1a** was a dibenzocyclooctadiene lignan possessing the same functional groups as schizandrin (**1b**) [2], namely, six methoxyl groups on the aromatic rings, a tertiary methyl group attached to a carbon carrying a hydroxyl group, a secondary methyl group and two benzylic methylenes.

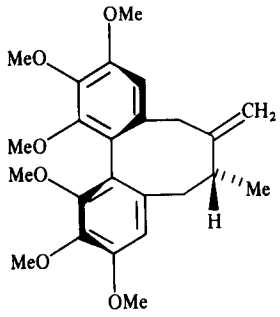
The constitution of **1a** was assumed by the ^1H–^{13}C shift correlated spectrum obtained by the correlation spectroscopy via long range coupling (COLOC) as shown in Fig. 1. The stereostructure of **1a** was elucidated by the measurements of the intramolecular nuclear Overhauser effect (NOE) in $CDCl_3$ as in the case of **1b**[2]. Irradiation of the methoxyl (δ3.89), H-9α (2.32) and tertiary methyl (1.19) groups caused 18, 12, 9% enhancements in the integrated intensity of the H-11 signal (6.62), respectively. On the other hand, no enhancement of the intensity of the H-4 signal (δ6.54) was detected upon irradiation of the C-7-secondary methyl signal (0.89). These findings indicate that the C-8 tertiary methyl group and H-11 are close to each other, while the C-7 secondary methyl group and H-4 are not. From these NOE data, the stereostructure of isoschizandrin was suggested as **1a** possessing a twist-boat–chair conformation of the cyclooctadiene ring.

The structure of **1a** including the biphenyl configuration was confirmed by the chemical correlation with (+)-deoxyschizandrin (**1c**) [3] and schizandrin (**1b**). Treatment of **1a** with phosphorus oxychloride in dry pyridine afforded **2**, $C_{24}H_{30}O_6$. The ^1H NMR spectrum of **2** in $CDCl_3$ showed two doublet signals (δ4.73 and 4.86, each *d*, J = 2 Hz) due to an exo-methylene group. Catalytic hydrogenation of **2** afforded (+)-deoxyschizandrin (**1c**), possessing a 7,8-*cis*-dimethyl group, and **1d**. A comparison of the ^1H NMR and ^{13}C NMR spectra of **1d** with those of *trans*-5,6,7,8-tetrahydro-1,12-dimethoxy-2,3:10,11-*bis* (methylenedioxy)-6,7-dimethylbenzo [*a*, *c*]-cyclooctene (**3**) synthesized by Tobinaga *et al.* [4] showed that **1d** possesses the same 7,8-*trans*-dimethyl group as **3**. The reaction mechanism requiring that hydrogenation of **2** occurs from the C-8α,17α side to afford **1d**, was confirmed by the finding that deuteration of **2** afforded **1e** (8α,17-dideutero compound of **1d**) and a minor compound which had a RR_t on HPLC the same as that of **1c**. By the chemical correlation of **1a** and **1c**, the configuration of the C-7 position in **1a** was determined to be *R* and that of the biphenyl group was also determined to be *R*.

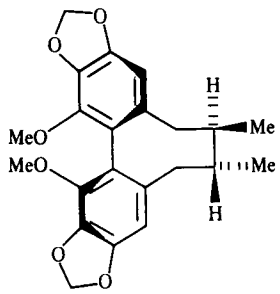
Finally, the configuration of the C-8 position in **1a** was confirmed by the transformation of schizandrin (**1b**) to **1a** as follows. Treatment of **1b** with phosphorous oxychloride in dry pyridine afforded a dehydrated product (**4a**) [5] which showed no hydroxyl band in the IR spectrum. The ^1H NMR spectrum of **4a** in $CDCl_3$ showed an olefinic proton signal (δ6.19, *br s*) and an olefinic methyl signal (δ1.63, *d*, J = 1.5 Hz), indicating the presence of the partial structure (Ar–CH=C(–)–Me). On oxidation with selenium dioxide in dioxane followed by acetylation **4a** afforded **4b**, $C_{24}H_{30}O_7$ and **4c**, $C_{26}H_{32}O_8$. The ^1H NMR spectrum of **4b** in C_6D_6 showed an olefinic methyl signal (δ1.76, *s*) and a tertiary methyl signal (1.47, *s*) attached to a carbon carrying a hydroxyl group (1.54), indicating that a hydroxyl group was attached to the C-8 carbon of **4a**. The configuration of the C-8 methyl group

* Part 14 in the series 'The Constituents of *Schizandra chinensis* Baill'; for Part 13 see ref. [1].



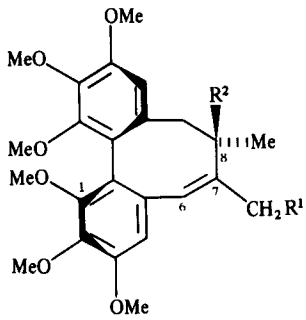


2



3

1a R^1 = H, R^2 = R^4 = Me, R^3 = OH
1b R^1 = OH, R^2 = R^4 = Me, R^3 = H
1c R^1 = R^3 = H, R^2 = R^4 = Me
1d R^1 = R^4 = H, R^2 = R^3 = Me
1e R^1 = H, R^2 = Me, R^3 = CH_2D, R^4 = D



4a R^1 = R^2 = H
4b R^1 = H, R^2 = OH
4c R^1 = OAc, R^2 = H

Table 1. ^{1}H NMR spectral data of compounds

Compound	H-4, *s* H-11, *s*	H-6α (*J* = Hz)	H-6β (*J* = Hz)	H-9α (*J* = Hz)	H-9β (*J* = Hz)
1a*	6.54	2.53, *d*(1.8)		2.32, *dd*	2.82, *d*
	6.52	2.54, *s*		(13.1, 0.8)	(13.1)
1a†	6.40	2.38, *dd*	2.63, *dd*	2.25, *d*	2.96, *d*
	6.48	(14, 8)	(14, 1.5)	(13)	(13)
1b	6.60	2.70, *d*	2.32, *d*	2.33, *dd*	2.68, *dd*
	6.53	(14)	(14)	(14, 7)	(14, 2)
1c‡	6.51	2.52 (centre)		2.27, *dd*	2.02, *dd*
	(× 2)	(2H, *m*)		(13.5, 9)	(13.5, 1)
1d	6.53	2.34, *dd*	2.53, *dd*	2.53, *dd*	2.34, *dd*
	(× 2)	(13.5, 7)	(13.5, 1.5)	(13.5, 1.5)	(13.5, 7)
1e	6.52	2.34, *dd*	2.53, *dd*	2.56, *d*	2.32, *d*
	(× 2)	(13.5, 7)	(13.5, 1.5)	(13.5)	(13.5)
3‡	6.48	2.25, *dd*	2.45, *dd*	2.45, *dd*	2.25, *dd*
	(× 2)	(13.4, 8)	(13.4, 2)	(13.4, 2)	(13.4, 8)

*This compound was measured at 400 MHz.
†This compound was measured in C_6D_6.

in **4b** was elucidated by measurements of the NOE in C_6D_6. Irradiation of the C-8 tertiary methyl group (δ1.47) and H-9α (2.65) caused 14 and 11% increases in the integrated intensity of the lower field aromatic proton signal (δ6.51, H-11), respectively. From this NOE data, the configuration of the C-8 position in **4b** was elucidated to be *R*. The presence of an acetoxymethyl signal (δ4.57, 1H, *d*, J = 13 Hz; 4.48, 1H, *d*, J = 13 Hz and 2.06, 3H, *s*) and no olefinic methyl signal in the ^{1}H NMR spectrum of **4c** indicated that the olefinic methyl group in **4a** changed to an acetoxymethyl group. Catalytic hydrogenation of **4b** gave a compound **1a** and it is assumed that **4b** was hydrogenated from the less hindered C-6β,7β side. From the above data, isoschizandrin was elucidated to have the formula **1a**.

Anti-ulcer activity was examined in water-immersed rats[6]. All samples were given orally 10 min before the water-immersion. As shown in Table 3, every sample inhibited the development of stress ulcer at 100 mg/kg, though their activities were weaker than that of schizandrin (**1b**).

EXPERIMENTAL

Mps: uncorr. ^{1}H NMR spectra were recorded at 60, 100, 200, and 400 MHz using TMS as int. standard. ^{13}C NMR spectra were recorded at 50 and 100 MHz using the same int. standard. UV spectra were measured in EtOH, IR spectra in KBr discs. Specific rotations were measured in $CHCl_3$. Kieselgel 60 (Merck) was used for CC and Kieselgel 60 F_{254} (Merck precoated plate) was used for TLC and prep. TLC. Spots were detected under UV (254 nm).

Isolation of isoschizandrin (**1a**). In the previous paper [2], it was reported that the petrol and MeOH extracts of the fruits of *Schizandra chinensis* Baill. (4.671 kg) afforded twelve fractions (frs 1–12) on silica gel CC with *n*-hexane, Me_2CO–benzene, and Me_2CO solvent systems. Fr. 10 (6.07 g) was chromatographed on silica gel (120 g) with an *n*-hexane–Me_2CO solvent system. The *n*-hexane–Me_2CO (4:1) eluate (1.623 g) was rechromatographed on silica gel (50 g) with an *n*-hexane–EtOAc solvent system. The *n*-hexane–EtOAc (73:27) and (70:30) eluates (1.153 g) were purified by repeated prep. TLC [1st: $CHCl_3$–EtOH (19:1), R_f 0.52; 2nd: *n*-hexane–EtOAc (1:2), R_f 0.51; 3rd: *n*-hexane–Me_2CO (3:2), R_f 0.35] to give isoschizandrin (**1a**) (56 mg, yield 0.0012%) as white amorphous powder, $[\alpha]_D^{25°}$ +92° ($CHCl_3$; *c* 1.22). IR ν_{max}^{KBr} cm^{-1}: 3480 (OH), 1595, 1575 (aromatic ring). $\nu_{max}^{CHCl_3}$ cm^{-1}: 3575 (OH), 1591, 1575 (aromatic). UV λ_{max}^{EtOH} nm (log ε): 218 (4.67), 251 (4.19), 284 (sh 3.38). MS *m/z* (rel. int.): 432 $[M]^+$ (2.8), 414 (100), 399 (9), 330 (14). High resolution MS *m/z*: 432.2198 (calc. for $C_{24}H_{32}O_7$: 432.2148).

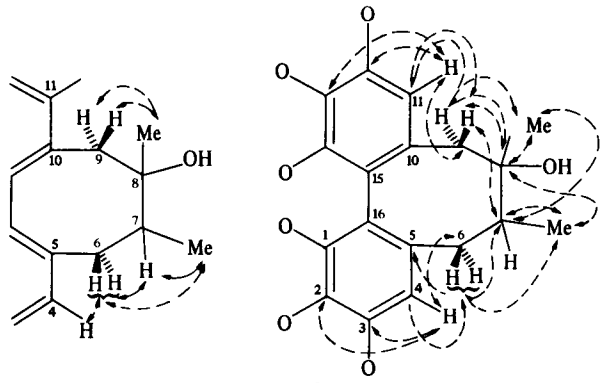
Dehydration of isoschizandrin (**1a**). Isoschizandrin (**1a**) (68 mg) was dissolved in a mixture of $POCl_3$ (0.4 ml) and dry pyridin (1 ml) and the soln heated at 70° for 1 hr. After cooling, the reaction mixture was diluted with Et_2O and poured into ice H_2O. The Et_2O soln was washed with H_2O, dried over Na_2SO_4 and evapd. The residue was purified by prep. TLC [*n*-hexane–EtOAc (7:3), R_f 0.6] to give a dehydrated compound (**2**) (36 mg) as colourless prisms from *n*-hexane, mp 115–117°, $[\alpha]_D^{25°}$ +198° (*c* 0.470, $CHCl_3$) (Found: C, 69.51; H, 7.33. $C_{24}H_{30}O_6$ requires: C, 69.54; H, 7.30). IR ν_{max}^{KBr} cm^{-1}: 1632 (C=C), 1596, 1576 (aromatic ring). MS *m/z* (rel. int.): 414 $[M]^+$ (100), 399 (11), 383 (6), 368 (8). ^{1}H NMR (200 MHz, $CDCl_3$): δ1.02 (3H, *d*, J = 7 Hz, H–C(–Me)), 2.53 (2H, *m*, $ArCH_2$–), 2.92 and 3.01 (each 1H, *d*, J = 12.5 Hz, $ArCH_2$–C=C), 2.68 (1H, *m*, –CH), 3.60 (3H, *s*), 3.62 (3H, *s*), 3.87 (3H, *s*), 3.90 (9H, *s*) (6 × OMe), 4.73 and 4.86 (each 1H, *d*, J = 2 Hz, –C=CH_2), 6.55 and 6.65 (each 1H, *s*, 2 × ArH).

Catalytic hydrogenation of **2**. Compound **2** (14 mg) in MeOH (2 ml) was hydrogenated in the presence of PtO_2 (25 mg) for 1.5 hr. The reaction mixture was purified by prep. HPLC *semi* prep. μ-Bondapak C_{18}, 7.8 mm i.d. × 30 cm; MeCN–MeOH–H_2O (10:10:12); 3.5 ml/min; UV 290 nm] to give **1c** (*RR*, 26.7 min) (2.5 mg) and **1d** (*RR*, 24.7 min) (5 mg). Compound **1c**. Colourless prisms from *n*-hexane–Et_2O; mp 114–116°; $[\alpha]_D^{24°}$ +80° (*c* 0.25, $CHCl_3$); IR ν_{max}^{KBr} cm^{-1}: 1591, 1576 (aromatic ring); MS *m/z* (rel. int.): 416 $[M]^+$ (100), 370 (6.2), 330 (5.2), 314 (4.8), 235 (6.9). This was identified as (+)-deoxyschizandrin by direct comparison (mmp, $[\alpha]_D$, IR, ^{1}H NMR) with an authentic sample. Compound **1d**. Colourless prisms from *n*-hexane; mp 84–86°; $[\alpha]_D^{24°}$ +82° (*c* 0.25, $CHCl_3$).

1a–1e, and **3** in $CDCl_3$ (200 MHz)

H-7	H-8	C-7-Me (J = Hz)	C-8-Me (J = Hz)	OMe *s*
1.89, *m*	1.78, *s* (OH)	0.89, *d* (7.1)	1.19, *s*	3.56, 3.57, 3.88 (× 2) 3.89 (× 2)
1.80, *m*	(OH)	0.92, *d* (7)	1.14, *s*	3.48, 3.49, 3.62 3.63, 3.85, 3.86
1.86, *s* (OH)	1.80, *m*	1.25, *s*	0.82, *d* (7)	3.59 (× 2), 3.90 (× 2) 3.92 (× 2)
	1.86 (centre) (2H, *m*)	1.00, *d* (7)	0.74, *d* (7)	3.58 (× 2), 3.88 (× 4)
	1.75 (centre) (2H, *m*)	0.86, *d* (7)	0.86, *d* (7)	3.56 (× 2), 3.88 (× 4)
1.75, *m*	—	0.85, *d* (7)	0.84, *s*	3.56 (× 2), 3.88 (× 4)
	1.72 (centre)	0.84, *d*	0.84, *d*	3.80 (× 2), 5.92 (1H, *d*, J = 1.22)
	(2H, *m*)	(7.3)	(7.3)	and 5.95 (1H, *d*, J = 1.22) (OCH_2O)

‡These compounds were measured at 100 MHz.



^{1}H - COSY(—) and ^{1}H - 2 - relayed COSY(····) spectra of **1a**

^{1}H–^{13}C shift correlated spectrum (····) of **1a** obtained by the COLOC

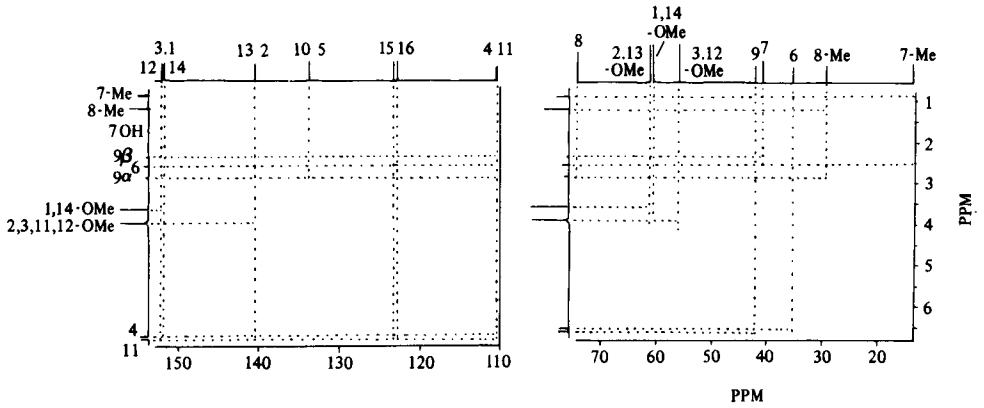


Fig. 1. Contour map of the ^{1}H–^{13}C shift correlated spectrum of 1a obtained by the COLOC (in $CDCl_3$). The J_{CH} parameter was set to 0.6 Hz.

(Found: C, 69.10; H, 7.74. $C_{24}H_{32}O_6$ requires: C, 69.21; H, 7.74); IR ν_{max}^{KBr} cm^{-1}: 1590, 1570 (aromatic ring); MS *m/z* (rel. int.); 416 $[M]^+$ (100), 370 (6), 314 (7).

Catalytic deuteration of **2**. Compound **2** (18 mg) in MeOH-d_4 (2 ml) was hydrogenated in the presence of PtO_2 (30 mg) for 1 hr. The reaction mixture was purified by prep. HPLC (as described above for the catalytic hydrogenation) to give a minor product (*RR*$_t$ 26.7 min) (1 mg) and **1e** (*RR*$_t$ 24.7 min) (12 mg) as a white amorphous powder; MS *m/z* (rel. int.): 419 $[M+1]^+$ (100), 418 $[M]^+$ (87), 402 (3); ^{1}H NMR: Table 1.

Dehydration of schizandrin (**1b**). Schizandrin (**1b**) (4 g) was dissolved in a mixture of $POCl_3$ (4 ml) and dry pyridine (20 ml), and the soln was heated at 90° for 2 hr. After cooling, the reaction mixture was diluted with Et_2O and poured into ice H_2O. The Et_2O soln was washed with H_2O, dried over Na_2SO_4 and evapd. The residue was recrystallized from *n*-hexane–Et_2O to afford a dehydrated compound (**4a**) (2.4 g) as colourless prisms, mp 122–122.5° (Found: C, 69.51; H, 7.39. Calc. for $C_{24}H_{30}O_6$: C, 69.54; H, 7.30). $[\alpha]_D^{25°}$ −97° ($CHCl_3$; *c* 1.07). This was identical (IR, ^{1}H NMR, MS, $[\alpha]_D$, mp) with an authentic sample [5].

Oxidation of **4a** *with selenium dioxide.* Compound **4a** (2.4 g) and selenium dioxide (1.2 g) were dissolved in dioxane (10 ml) and the soln was stirred for 6 hr at 45°. The reaction mixture was diluted with Et_2O, washed with H_2O, dried over Na_2SO_4 and evapd. The residue was chromatographed on silica gel (3 cm i.d. × 20 cm) with an *n*-hexane–EtOAc solvent system. The fractions eluted with *n*-hexane–EtOAc (60:40) were combined and evapd to give a residue, which was acetylated by the mixture of Ac_2O (1 ml) and dry pyridine (2 ml). The reaction mixture was purified by prep. TLC [*n*-hexane–Me_2CO (7:3)] to give **4b** (R_f 0.3, 285 mg) and **4c** (R_f 0.5, 410 mg). Compound **4b**. Colourless prisms from *n*-hexane–Et_2O; mp 178–180°; $[\alpha]_D^{23°}$ −41° ($CHCl_3$; *c* 2.36) (Found: C, 66.95; H, 7.18. $C_{24}H_{30}O_7$ requires: C, 66.96; H, 7.02). IR ν_{max}^{KBr} cm^{-1}: 3480 (OH), 1592, 1579 (aromatic ring); UV λ_{max}^{EtOH} nm (log ε): 211 (4.54), 228 (4.55), 294 (sh 3.33); MS *m/z* (rel. int.): 430 $[M]^+$ (100), 412 (33), 399 (11); ^{1}H NMR (200 MHz, C_6D_6): δ1.47 (3H, *s*, HO–$\overset{|}{\underset{|}{C}}$–Me), 1.54 (1H, *br s*, OH), 1.76 (3H, *s*, HC=$\overset{|}{\underset{|}{C}}$–Me), 2.65 (1H, *d*, *J* = 13 Hz), 3.24 (1H, *d*, *J* = 13 Hz) ($ArCH_2$–), 3.44 (3H, *s*), 3.57 (3H, *s*), 3.62 (3H, *s*), 3.72 (3H, *s*), 3.78 (3H, *s*), 3.85 (3H, *s*) (6 × OMe), 6.28 (1H, *br s*, HC=$\overset{|}{\underset{|}{C}}$–Me), 6.45 (1H, *s*, H-4), 6.51 (1H, *s*, H-11). Compound **4c** yellow oil, $[\alpha]_D^{23°}$ −178° ($CHCl_3$; *c* 1.76). $C_{26}H_{32}O_8$ (high resolution MS: 472.2150; Calc. 472.2097); IR ν_{max}^{KBr} cm^{-1}: 1738 (C=O), 1594, 1574 (aromatic ring); UV λ_{max}^{EtOH} nm (log ε): 211 (4.57), 242 (sh 4.35), 288 (sh 3.39); MS *m/z* (rel. int.): 472 $[M]^+$ (100), 412 (5), 381 (24), 300 (18); ^{1}H NMR (200 MHz, $CDCl_3$): δ1.12 (3H, *d*, *J* = 7 Hz, –$\overset{|}{C}$H–Me), 2.06 (3H, *s*, COMe), 2.36 (1H, *dd*, *J* = 14, 8 Hz), 3.14 (1H, *dd*, *J* = 14, 7.5 Hz) ($ArCH_2$–), 2.88 (1H, *m*–$\overset{|}{C}$H), 3.53 (3H, *s*), 3.64 (3H, *s*), 3.86 (3H, *s*), 3.88 (3H, *s*), 3.90 (3H, *s*) (6 × OMe), 4.48 (1H, *d*, *J* = 13 Hz), 4.54 (1H, *d*, *J* = 13 Hz) (=$\overset{|}{C}$–CH_2OCOMe), 6.43 (1H, *s*, ArH), 6.46 (1H, *s*, ArH), 6.50 (1H, *br s*, HC=$\overset{|}{\underset{|}{C}}$–).

Catalytic hydrogenation of **4b**. Compound **4b** (160 mg) in MeOH (3 ml) was hydrogenated in the presence of 10% Pd-C (160 mg) for 7 hr. The reaction mixture was purified by prep. TLC

Table 2. ^{13}C NMR spectral data of compounds **1a–1d**, and **3** (δ in $CDCl_3$, 50 MHz)

C		1a*	1b	1c	1d	3
1		151.7	151.9[a]	151.6[a]	151.6	141.2
2		140.3	140.8[b]	140.3[b]	140.2	134.9
3		151.8	152.3	153.0	151.6	147.5
4		110.6	110.5[c]	107.3	110.7	106.1
5		133.65	131.8	139.1	134.2	133.0
6		35.4	40.9	35.7	33.1	32.8
7		40.8	71.8	40.9	36.0	35.9
8		74.1	41.8	33.8	36.0	35.9
9		42.1	34.4	39.2	33.1	32.9
10		133.67	133.8	133.9	134.2	133.0
11		110.4	110.1[c]	110.6	110.6	106.1
12		152.0	152.0	151.7	151.6	147.5
13		140.5	140.3[b]	139.9[b]	140.2	134.9
14		151.7	151.6[a]	151.5[a]	151.6	141.2
15		123.3	122.8	123.5	123.7	122.4
16		122.9	124.2	122.4	123.7	122.4
17		29.2	15.9	12.7	20.4	20.5
18		13.5	29.7	21.8	20.4	20.5
	C-1,14	60.5 (×2)	60.6 (×2)	60.3 (×2)	60.5 (×2)	59.6 (×2)
OMe	C-2,13	60.9 (×2)	61.0 (×2)	60.7 (×2)	60.9 (×2)	—
	C-3,12	56.0 (×2)	55.9 (×2)	55.7 (×2)	56.0 (×2)	—
OCH_2O		—	—	—	—	100.7

*This compound was measured at 100 MHz and assignments for the carbon signals were confirmed by the ^1H-^{13}C shift correlation 2D-NMR spectrum.

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

Table 3. Effect of schizandrin and its derivatives on water-immersion stress-induced gastric ulcer in rats

Compound	Dose (mg/kg, p.o.)	Number of rats	Inhibitory ratio (%)
control		5	—
1a	100	5	39.9
1b	100	5	52.5
2	100	5	34.1
1c	100	5	30.7
4a	100	5	20.7
4b	100	5	15.9
4c	100	5	25.1
cimetidine	50	5	61.5

$[CHCl_3$–EtOH (20:1), R_f 0.45] to give product **1a** (79 mg) as a white amorphous powder, $[\alpha]_D^{23}$ +90° ($CHCl_3$; *c*0.730) (Found: C, 66.40; H, 7.33. $C_{24}H_{32}O_7$ requires: C, 66.65; H, 7.46). This was identified as isoschizandrin by direct comparison ($[\alpha]_D$, IR, ^1H NMR, ^{13}C NMR) with an authentic sample.

Bioassay. Male Wister strain rats weighing 230–250 g were placed in a stress cage and immersed to the level of the xiphoid process in a water bath (23°) for 7 hr. The animals were then immediately sacrificed by a blow on the head. The stomach of each was removed, slightly inflated by injecting 1% formaline soln for 10 min to flex the inner and outer layers of gastric walls. The stomach was then incised along the greater curvature and examined for lesions developed in the glandular portion. The ulcer index was calculated as the sum of the length (mm) of lesions. Samples were suspended in 1% Tween 80 soln. Cimetidine (100 mg/kg or 50 mg/kg) as a positive control was given orally 10 min before the water-immersion.

Inhibitory ratio (%)

$$= \frac{\text{ulcer index (control)} - \text{ulcer index (sample)}}{\text{ulcer index (control)}}$$

Acknowledgements—We thank Dr M. R. Wälchli, Bruker Japan Co. Ltd, for measurements of 400 MHz NMR spectra, and Mr H. Kobayashi, Faculty of Pharmaceutical Science, University of Tokyo, for measurement of NOE. We also thank Mr K. Sato, Meiji College of Pharmacy for elemental analysis, and Mr I. Sakakibara, Tsumura Laboratory for measurements of mass spectra.

REFERENCES

1. Nakajima, K., Taguchi, H., Ikeya, Y., Endo, T. and Yosioka, I. (1983) *Yakugaku Zasshi* **103**, 743.
2. Ikeya, Y., Taguchi, H., Yosioka, I. and Kobayashi, H. (1979) *Chem. Pharm. Bull.* **27**, 1383.
3. Ikeya, Y., Taguchi, H., Yosioka, I. and Kobayashi, H. (1979) *Chem. Pharm. Bull.* **27**, 2695.
4. Takeya, T., Okubo, T., Nishida, S. and Tobinaga, S. (1985) *Chem. Pharm. Bull.* **33**, 3599.
5. Ikeya, Y., Taguchi, H., Sasaki, H., Nakajima, K. and Yosioka, I. (1980) *Chem. Pharm. Bull.* **28**, 2414.
6. Takagi,K. and Okabe, S. (1968) *Jpn J. Pharmacol.* **18**, 9.