Chemical Conversion of Vibsanin C to Vibsanin E and Structure of 3-Hydroxyvibsanin E from *Viburnum awabuki*

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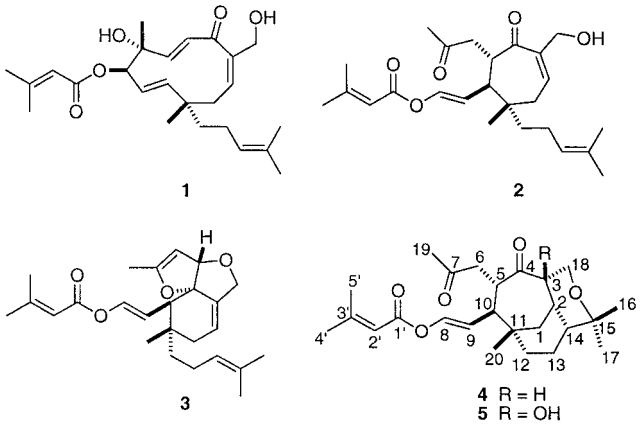
Received August 7, 1998

Vibsanin E (**4**), a tricyclic vibsane-type diterpene, has been prepared in 50% yield from vibsanin C (**2**), a seven-membered ring vibsane-type diterpene by reaction with BF_3·OEt_2 at −78 °C. This chemical correlation not only established structure, including absolute configurations, but also has demonstrated a possible biosynthetic route to **4** via **2** derived from vibsanin B (**1**). The structure of 3-hydroxyvibsanin E (**5**), another example of a tricyclic seven-membered ring vibsane, isolated from the leaves of *Viburnum awabuki*, has been established by extensive analyses of 2D NMR data and comparison of its spectral data with those of **4**.

Vibsane-type diterpenes,[1] which consist of a fumulane-type carbon skeleton with an additional isoprene unit, are very rare in nature. Their occurrence had been limited to the genus *Viburnum* (Caprifoliaceae)[2] until some examples of *ent*-vibsane-type diterpenes were isolated from the liverwort *Odontoschisma denudantum*.[3] The carbon skeletons of vibsane-type diterpenes can be classified into three subgroups characterized by an eleven-membered ring, a seven-membered ring, and a rearranged type and are represented by vibsanin B (**1**), vibsanin C (**2**),[4] and neovibsanin (**3**),[5] respectively. In previous papers,[1,5] we successfully correlated **1** to **2** and **3** by thermal and photochemical reactions, respectively, and thereby not only established their absolute configurations but also suggested a plausible biosynthesis for the three subtypes of vibsane-type diterpenes. Another vibsane-type diterpene, vibsanin E (**4**), was isolated from *V. awabuki* by K. Kawazu, and its absolute structure was unambiguously established by X-ray crystallographic analysis.[6] One assumes that the unique tricyclic structure of **4** must be formed somehow from **2**, but a chemical method to construct the tricyclic system in **4** had not been achieved. In this paper, we report the successful chemical conversion of **2** to **4** and the structure elucidation of a new diterpene, 3-hydroxyvibsanin E (**5**), isolated from the leaves of *V. awabuki*. The leaves of *V. awabuki* K. Koch (Caprifoliaceae) were extracted with MeOH, and the evaporated extract was purified by repeated chromatography on Si gel to yield **4** and **5**.

The initial attempts to convert **2** to **4** were unsuccessful. Although treatment of **2** with acids was found to give some tricyclic products, the acid-sensitive enol ester group in **2** caused side reactions and yielded no product corresponding to **4**. After several efforts failed, we found that when **2** was treated with boron trifluoride diethyl etherate in CH_2Cl_2 under anhydrous conditions at −78 °C for a short period, it gave **4** in 50% yield.

Compound **5** was obtained as a colorless, amorphous solid. Its FABMS exhibited quasi-molecular ion peaks at *m*/*z* 433 and 455, and the molecular formula $C_{25}H_{36}O_6$, consistent with eight unsaturation units, was assigned by HRFABMS. Absorption bands in the IR (3461, 1728, and 1710 cm^{-1}) and UV (230 nm) spectra of **5** suggested the presence of a hydroxyl, carbonyl, and conjugated ester groups. The ^1H and ^{13}C NMR data (Table 1) of **5** showed the presence of six tertiary methyl groups (δ 0.82, 1.13, 1.18, 1.37, 1.38, and 2.05), an isolated oxymethylene (δ 3.66, 4.36; δ 65.9), a trisubstituted olefin (δ 5.70; δ 115.0, 160.0), and a disubstituted olefin (δ 4.99, 7.28; δ 115.8, 135.2). These data were very similar to those of **4**. Comparison of the NMR data (Table 1) of **5** and **4** indicated that **5** had the hydroxy group at the C-3 position in **4**. This hydroxyl proton signal, which was assigned to δ 4.91 (s), showed distinct HMBC correlations to C-2, C-3, C-4, and C-18, thereby supporting its position on C-3. The other partial structures obtained by 2D COSY and HMQC and their assembly made up by HMBC as shown in Table 1 indicated that **5** is 3-hydroxyvibsanin E. The relative stereochemistry for **5** was assigned to be the same as that of **4** on the basis of cross peaks as shown in Figure 1 on 2D NOESY. The CD spectrum of compound **5** displayed the same positive Cotton effect as that of **4**. Therefore, 3-hydroxyvibsanin E (**5**) had 3*S*, 5*S*, 10*R*, and 11*S* configurations.



The present study has demonstrated that vibsanin E (**4**), the first example of a tricyclic seven-membered ring vibsane, can be derived from the seven-membered ring vibsane, vibsanin C (**2**) by a cationic process and has established that **4** has the same absolute configuration as **2**. From a biosynthetic point of view, **4** may be formed in the plant from **2** available from the eleven-membered ring vibsane, vibsanin B (**1**). 3-Hydroxyvibsanin (**5**) is the second example of a tricyclic seven-membered ring vibsane. This structural diversity of the vibsane-type diterpenes encour-

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10.1021/np980338v CCC: $18.00
Published on Web 12/05/1998

Table 1. NMR Spectra Data of Compounds **4** and **5** in C_6H_6-d_6 (600 MHz for ^1H NMR, 150 MHz for ^{13}C NMR)

	4		**5**		
position	δ_H[a]	δ_C[b]	δ_H[a]	δ_C[b]	HMBC Correlations[c]
1	1.10 (dd, 14.8, 5.5)	41.3	1.03 (dd, 14.8, 5.5)	36.6	C-2, C-3, C-10, C-11,
	2.00 (br d, 14.8)		2.68 (dd, 14.8, 2.1)		C-2, C-11, C-12, C-14
2	2.13 (m)	29.8	2.41 (ddd, 5.5, 4.4, 2.5)	37.1	C-3, C-4, C-11, C-14, C-15
3	2.26 (dd, 7.7, 3.3)	50.1		74.4	
4		212.9		213.8	
5	3.06 (ddd,11.5,10.7, 3.0)	47.7	3.43 (ddd, 11.8, 11.8, 3.0)	43.1	C-10
6	2.13 (dd, 17.6, 3.0)	46.1	2.01 (dd, 19.0, 3.0)	46.4	C-4, C-7, C-10
	2.98 (dd, 17.6, 10.7)		2.91 (dd, 19.0, 11.8)		C-4, C-7, C-10
7		206.9		210.8	
8	7.25 (d, 12.4)	135.1	7.28 (d, 12.4)	135.2	C-1′, C-9, C-10
9	5.09 (dd, 12.4, 11.8)	116.3	4.99 (dd, 12.4, 11.8)	115.8	C-8
10	1.72 (dd, 11.8, 11.5)	46.0	1.68 (t, 11.8)	45.7	C-4, C-5, C-8, C-9, C-11, C-12
11		34.9		35.3	
12	0.76 (ddd, 14.1, 14.1, 3.3)	42.4	0.77 (m)	42.0	
	1.25 (m)		1.25 (m)		
13	1.16 (m), 1.25 (m)	19.1	1.16 (m), 1.29 (m)	19.3	C-14
14	0.67 (ddd, 12.6, 4.4, 4.1)	43.4	0.92 (ddd, 13.2, 4.4, 4.4)	43.9	
15		72.5		72.7	
16	1.11 (s)	23.3	1.18 (s)	23.5	C-14, C-15, C-17
17	1.04 (s)	28.3	1.13 (s)	27.8	C-14, C-15, C-16
18	3.32 (dd, 11.8, 3.3)	59.2	3.66 (d, 11.4)	65.9	C-2, C-3, C-4, C-15
	4.48 (d, 11.8)		4.36 (d, 11.4)		C-3, C-4
19	1.62 (s)	29.5	1.38 (s)	28.9	C-6, C-7
20	0.79 (S)	31.9	0.82 (s)	31.7	C-1, C-10, C-11, C-12
1′		163.2		163.2	
2′	5.69 (qq, 1.1, 1.1)	115.1	5.70 (qq, 1.1, 1.1)	115.0	C-4′, C-5′
3′		159.8		160.0	
4′	1.37 (d, 1.1)	20.3	1.37 (d, 1.1)	20.3	C-2′, C-3′, C-5′
5	2.06 (d, 1.1)	27.3	2.05 (d, 1.1)	27.0	C-2′, C-3′, C-4′
C_3−OH			4.91 (s)		C-2, C-3, C-4, C-18

[a] Chemical shift (δ) in ppm from TMS, multiplicities, and coupling constants in Hz are in parentheses. [b] Chemical shift (δ) in ppm from TMS. [c] Correlations from H to the indicated carbons.

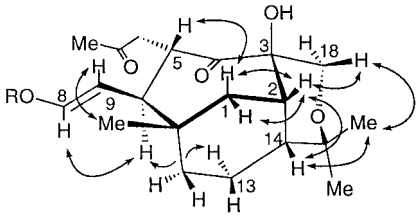

Figure 1. Relative stereochemistry for **5** based on NOESY. Arrows indicate NOE correlations.

ages us to continue studying the chemical components of *Viburnum* species.

Experimental Section

General Experimental Procedures. Melting points were determined with a Mitamura hot-stage microscope and are uncorrected. IR and UV spectra were obtained using a JASCO FTIR-5300 and a Shimazu UV-300 spectrophotometer, respectively. NMR spectra were recorded on a Varian Unity 600 and 200, with TMS as the internal standard. The HMBC, HMQC, and ROESY were run on a Varian Unity at 600 MHz. MS were measured with a JEOL JMS AX-500. CD spectra were recorded on a JASCO J-500. Si gel 60 (Merck) and Wako gel C-300 (Wako) were used for column chromatography. Analytical TLC was carried out on Si gel plates (Kieselgel 60, Merck) and visualized by 5% $CeSO_4$ in H_2SO_4 followed by heating.

Plant Material. The leaves of *Viburnum awabuki* were collected in Tokushima, Japan. A voucher specimen (1315LF) has been deposited at the Institute of Pharmacognosy, Tokushima Bunri University.

Extraction and Isolation. The dried and powdered leaves of *V. awabuki* (1.5 kg) were immersed in MeOH at room temperature for 1 month. The MeOH extract was evaporated in vacuo to give a gummy extract (500 g). The extract (20 g) was subjected to a Si gel (Merck) column chromatography eluted, in order, with hexane (100%), hexane−EtOAc (7:3), hexane−EtOAc (1:1), EtOAc (100%), EtOAc−MeOH (8:2), and MeOH (100%), yielding 10 fractions. Fraction 5 (4.9 g) was rechromatographed on Si gel (Wako) with $CHCl_3$−EtOAc (5:1) to give fractions 11−16. Fraction 13 (1.12 g) was subjected to reversed-phase chromatography using Cosmosil 75C18−OPN and eluted with MeOH−H_2O (5:1) to give fractions 17−19. Fraction 18 (66 mg) was purified by Si gel chromatography with hexane−EtOAc (2:1) to afford vibsanin E (**4**) (11.1 mg) and 3-hydroxyvibsanin E (**5**) (12.8 mg).

Vibsanin E (4): colorless prisms, mp 147−148 °C; $[\alpha]^{21}_D$ −61° (*c* 0.3, $CHCl_3$); IR ($CHCl_3$) 1732, 1647 cm^{-1}; UV λ_{max} (EtOH) 230 (ϵ 13 700) nm; CD (EtOH) $\Delta\epsilon_{275}$ +15.2; ^1H and ^{13}C NMR, see Table 1; FABMS *m*/*z* 439 $[M + Na]^+$, 417 $[M + H]^+$; HRFABMS *m*/*z* 439.2468 $[M + Na]^+$ (calcd for $C_{25}H_{36}O_5Na$, 439.2461).

3-Hydroxyvibsanin E (5): colorless amorphous solid; $[\alpha]^{21}_D$ +6.3° (*c* 0.3, EtOH); IR (neat) 3461, 1728, 1710 cm^{-1}; UV λ_{max} (EtOH) 230 (ϵ 10 000) nm; CD (EtOH) $\Delta\epsilon_{278}$ +15.7; ^1H and ^{13}C NMR, see Table 1; FABMS *m*/*z* 455 $[M + Na]^+$, 433 $[M + 1]^+$; HRFABMS *m*/*z* 455.2443 $[M + Na]^+$, (calcd for $C_{25}H_{36}O_6Na$, 455.2410).

Conversion of Vibsanin C (2) to Vibsanin E (4): To a solution of vibsanin C (**2**) (8 mg) in CH_2Cl_2 (0.5 mL) was added boron trifluoride diethyl etherate (0.025 mL) at −78 °C. After being stirred for 18 min, the mixture was diluted with a saturated aqueous solution of $NaHCO_3$ and extracted with Et_2O and washed with H_2O and brine, then dried over $MgSO_4$. The solvent was removed in vacuo to give a crude oil, which was purified by HPLC [Cosmosil 5C18 AR, ϕ10 × 250 mm; solvent: MeOH−MeCN−H_2O (3:1:1.5), 2 mL/min] to yield vibsanin E (**4**) (4 mg, 50%) as colorless prisms: 147−148 °C; $[\alpha]^{21}_D$ −65° (*c* 0.3, $CHCl_3$). All spectral data of synthetic **4** were identical with those of the natural product isolated from *V. awabuki*.

Acknowledgment. We thank Mr. S. Takoka and Mrs. I. Okamoto of the Instrument Analysis Center for Pharmaceutical Sciences, Tokushima Bunri University, for MS and NMR measurements. This work was partially supported by a Grant-

in-Aid for Scientific Research (no. 09680582) from the Ministry of Education, Science, Sport and Culture, Japan.

References and Notes

(1) Fukuyama, Y.; Minami, H.; Takaoka, S.; Kodama, M.; Kawazu, K.; Nemoto, H. *Tetrahedron Lett*. **1997**, *38*, 1435−1438.
(2) Connelly, J. D.; Hill, R. A. In *Dictionary of Terpenoids*; Chapman & Hall: London, 1991; Vol. 2, pp 1084−1085.
(3) Hashimoto, T.; Toyota, M.; Koyama, H.; Kikkawa, A.; Yoshida, M.; Tanaka, M. Takaoka, S.; Askawa, Y. *Tetrahedron Lett*. **1998**, *39*, 579−582.
(4) Kawazu, K. *Agric. Biol. Chem*. **1980**, *44*, 1367−1372.
(5) Fukuyama, Y.; Minami, H.; Takeuchi, K.; Kodama M.; Kawazu K. *Tetrahedron Lett*. **1996**, *37*, 6767−6770.
(6) Fukuyama, K.; Katsube, Y.; Kawazu, K. *J. Chem. Soc., Perkin 2* **1980**, 1701−1703.

NP980338V