Crystal Structures of 5α,6α-Epoxy and 2,3-Dihydro Derivatives of Physalin B, a 13,14-Seco-16,24-cyclosteroid, and Their ^{1}H NMR Spectral Analysis

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X-Ray crystallographic analysis of two steroid derivatives, 5α,6α-epoxyphysalin B and 2,3-dihydrophysalin B, have been undertaken, revealing their closely similar crystal structures to each other. Their ^{1}H NMR spectra, as well as that of the parent compound physalin B, a 13,14-seco-16,24-cyclosteroid from *Physalis alkekengi*, have been analyzed based on the crystal structures of these derivatives.

Physalins are the steroidal constituents of *Physalis* plants (*Solanaceae*) possessing a novel 13,14-seco-16,24-cycloergostane skeleton. Since the isolation and structure determination of physalin A (**1**) and physalin B (**2**) from *P. alkekengi* var. *francheti*,[1] more than a dozen physalins were isolated from Japanese and Indian *Physalis* plants.[2] In addition to the novel skeletal structure carrying many functional groups, the *in vitro* and *in vivo* antitumor activity found for **2** and (25*S*)-25,27-dihydro-7-dehydroxyphysalin A[3] makes the physalins an interesting family of natural products. Knowledge of their accurate geometry is essential for understanding the structure–antitumor activity relationships of physalins and related compounds. X-Ray crystallographic and NMR spectroscopic analyses are the most useful techniques for the structural study. An X-ray analysis of a hydrogenated derivative of **1**, (25*R*)-2,3,25,27-tetrahydrophysalin A (**3**) and the ^{1}H NMR spectral analysis of **1** based on the crystal structure of **3**, were already reported.[4] While **1** is heptacyclic, **2** possesses an octacyclic structure containing an additional intramolecular acetalic linkage C(14)–O–C(27) (Chart 1). Since crystallographic analysis of **2** and related physalins has not yet been reported, X-ray analysis of **2** and its derivatives has been attempted. Although **2** itself did not afford crystals suitable for crystallography, the 5α,6α-epoxy derivative **4**, known as physalin J,[5,6] and the 2,3-dihydro derivative **5**[1] were found to give well-grown prismatic crystals which could be subjected to crystallographic analysis. This paper describes the crystal structures of **4** and **5** and the ^{1}H NMR spectral analysis of **2**, **4**, and **5** based on their crystal structures.

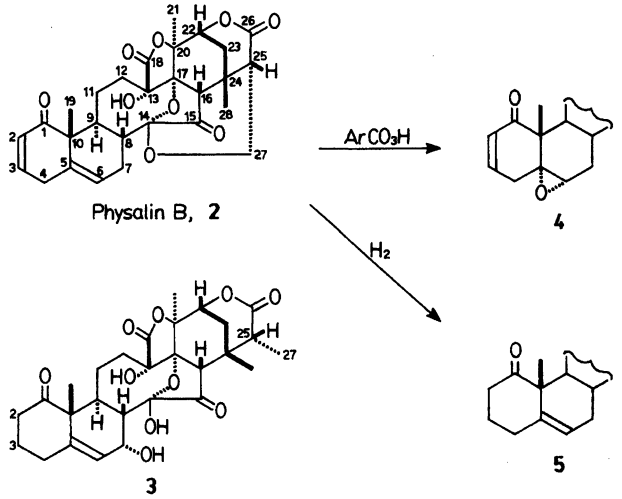

Chart 1.

Experimental

Mp's were determined on a hot plate apparatus and are uncorrected. 400 MHz ^{1}H NMR spectra were recorded in dimethyl-d_6 sulfoxide solutions at 27 °C on a JEOL JNM-GSX 400 spectrometer.

Physalin B (2). This compound was isolated from *P. alkekengi* var. *francheti* as described previously.[1] Mp 246—248 °C from acetone (lit,[1] mp 245—250 °C).

5α,6α-Epoxyphysalin B (4). A $CHCl_3$ solution (10

Table 1. Crystal Data and Experimental Details for **4** and **5**

	4	**5**
Formula	$C_{28}H_{30}O_{10}$	$C_{28}H_{28}O_9$
Formula weight	526.54	512.55
Crystal system	Monoclinic	Monoclinic
Space group	$P2_1$	$P2_1$
a/Å	10.286(2)	10.381(3)
b/Å	12.010(4)	11.842(3)
c/Å	9.564(2)	9.540(5)
β/°	91.81(2)	91.99(2)
V/Å^3	1180.9	1172.1
Z	2	2
F(000)	556.0	544.0
μ(Cu $K\alpha$)/cm^{-1}	8.987	8.557
Crystal size/mm^3	0.4×0.4×0.3	0.3×0.3×0.2
Scan mode	2θ–ω	2θ–ω
Scan width		
2θ range/°	<120	<120
Number of reflections ($F_o>3\sigma$)	1880	1831
R	0.0515	0.0412
wR	0.0670	0.0591

Table 2. Fractional Coordinates and Equivalent Isotropic Temperature Factors[a] of Non-hydrogen Atoms[b] of **4** and **5**

	4				**5**			
Atom	x	y	z	U_{eq}	x	y	z	U_{eq}
C(1)	0.9936(3)	0.6798(3)	1.0876(4)	0.038(1)	1.0048(3)	0.6750(3)	1.1018(3)	0.037(1)
C(2)	1.0825(4)	0.6809(4)	1.2117(4)	0.052(1)	1.0954(4)	0.6853(4)	1.2262(4)	0.048(1)
C(3)	1.1993(4)	0.6320(5)	1.2084(4)	0.059(2)	1.1837(4)	0.5823(4)	1.2313(4)	0.061(1)
C(4)	1.2497(4)	0.5753(4)	1.0838(4)	0.052(1)	1.2513(3)	0.5655(4)	1.0955(4)	0.054(1)
C(5)	1.1423(3)	0.5612(4)	0.9702(4)	0.040(1)	1.1552(3)	0.5602(3)	0.9693(4)	0.042(1)
C(6)	1.1548(3)	0.4750(4)	0.8655(4)	0.046(1)	1.1582(3)	0.4779(4)	0.8772(4)	0.045(1)
C(7)	1.0849(4)	0.4811(4)	0.7262(5)	0.054(1)	1.0686(3)	0.4666(4)	0.7524(4)	0.046(1)
C(8)	0.9948(3)	0.5821(3)	0.6996(4)	0.037(1)	0.9943(3)	0.5745(3)	0.7150(3)	0.036(1)
C(9)	0.9462(3)	0.6372(3)	0.8351(3)	0.032(1)	0.9496(3)	0.6357(3)	0.8465(3)	0.032(1)
C(10)	1.0560(3)	0.6613(3)	0.9463(3)	0.032(1)	1.0628(3)	0.6590(3)	0.9556(3)	0.033(1)
C(11)	0.8705(3)	0.7465(3)	0.8019(4)	0.035(1)	0.8754(3)	0.7470(3)	0.8178(3)	0.035(1)
C(12)	0.7198(3)	0.7458(3)	0.7889(4)	0.036(1)	0.7266(3)	0.7493(3)	0.8014(3)	0.038(1)
C(13)	0.6505(3)	0.6359(4)	0.8034(3)	0.035(1)	0.6536(3)	0.6390(3)	0.8134(3)	0.036(1)
C(14)	0.8785(3)	0.5446(3)	0.6049(3)	0.033(1)	0.8788(3)	0.5420(3)	0.6192(3)	0.035(1)
C(15)	0.8007(3)	0.6366(3)	0.5308(3)	0.034(1)	0.8056(3)	0.6374(3)	0.5430(3)	0.035(1)
C(16)	0.6590(3)	0.6069(3)	0.5414(3)	0.035(1)	0.6634(3)	0.6100(3)	0.5510(3)	0.033(1)
C(17)	0.6641(3)	0.5476(3)	0.6850(3)	0.033(1)	0.6651(3)	0.5492(3)	0.6954(3)	0.033(1)
C(18)	0.5014(3)	0.6486(4)	0.7909(4)	0.043(1)	0.5069(3)	0.6558(4)	0.8003(3)	0.043(1)
C(19)	1.1411(3)	0.7614(4)	0.9094(4)	0.037(1)	1.1420(3)	0.7655(4)	0.9172(3)	0.044(1)
C(20)	0.5464(3)	0.4709(4)	0.7090(4)	0.039(1)	0.5452(3)	0.4745(3)	0.7160(3)	0.039(1)
C(21)	0.5654(4)	0.3860(4)	0.8277(4)	0.051(1)	0.5582(4)	0.3877(4)	0.8353(4)	0.050(1)
C(22)	0.4927(3)	0.4131(4)	0.5761(4)	0.042(1)	0.4903(3)	0.4181(4)	0.5812(4)	0.045(1)
C(23)	0.4816(3)	0.4863(4)	0.4505(4)	0.043(1)	0.4843(3)	0.4935(4)	0.4560(3)	0.044(1)
C(24)	0.6150(3)	0.5314(3)	0.4160(4)	0.039(1)	0.6184(3)	0.5352(3)	0.4228(3)	0.039(1)
C(25)	0.7043(4)	0.4309(4)	0.3911(4)	0.040(1)	0.7048(3)	0.4308(3)	0.3980(3)	0.041(1)
C(26)	0.6843(4)	0.3274(3)	0.4777(4)	0.042(1)	0.6789(3)	0.3261(3)	0.4829(3)	0.043(1)
C(27)	0.8505(4)	0.4548(4)	0.3782(4)	0.047(1)	0.8497(3)	0.4522(4)	0.3875(3)	0.046(1)
C(28)	0.6052(4)	0.6034(4)	0.2855(4)	0.047(1)	0.6146(3)	0.6087(4)	0.2931(3)	0.047(1)
O(1)	0.8764(2)	0.6903(3)	1.1014(3)	0.048(1)	0.8893(2)	0.6731(3)	1.1175(2)	0.051(1)
O(5)	1.0725(3)	0.4575(3)	0.9833(3)	0.053(1)				
O(13)	0.6734(2)	0.5891(3)	0.9382(2)	0.042(1)	0.6751(2)	0.5895(2)	0.9477(2)	0.043(1)
O(14)	0.9255(2)	0.4716(3)	0.5059(3)	0.043(1)	0.9237(2)	0.4656(2)	0.5190(2)	0.042(1)
O(15)	0.8464(3)	0.7147(3)	0.4696(3)	0.047(1)	0.8537(2)	0.7153(2)	0.4833(2)	0.049(1)
O(17)	0.7836(2)	0.4868(2)	0.6865(2)	0.032(1)	0.7816(2)	0.4848(2)	0.6986(2)	0.032(1)
O(18)	0.4382(3)	0.7285(3)	0.8198(3)	0.059(1)	0.4464(2)	0.7377(3)	0.8291(3)	0.064(1)
O(20)	0.4471(2)	0.5522(3)	0.7456(3)	0.045(1)	0.4491(2)	0.5597(3)	0.7506(2)	0.046(1)
O(22)	0.5738(3)	0.3168(3)	0.5497(3)	0.048(1)	0.5688(3)	0.3184(2)	0.5562(3)	0.050(1)
O(26)	0.7555(3)	0.2488(3)	0.4763(3)	0.062(1)	0.7463(3)	0.2436(3)	0.4810(3)	0.062(1)

a) $U_{eq}=(1/3)\sum_i\sum_j U_{ij}a_i^*a_j^*\boldsymbol{a}_i\cdot\boldsymbol{a}_j$. b) The oxygen atom is assigned the same number as its bonded carbon atom for ease of comparing the two structures.

cm^3) containing *m*-chloroperbenzoic acid (0.86 mmol) was added portionwise to an ice-cooled solution of **2** (364 mg, 0.71 mmol) in $CHCl_3$ (30 cm^3) and the mixture was stirred at room temperature for 29 h. The crude product composed of 5α,6α-epoxide (**4**) and 5β,6β-epoxide (physalin F[5,6]) was subjected to repeated silica-gel column chromatography using $CHCl_3$–MeOH and benzene–AcOEt as eluents to give pure **4** (148 mg, yield 39%). Crystallization from MeOH–acetone (1:1) afforded colorless prisms, mp 272.5—274 °C (lit,[6] 268—270 °C).

2,3-Dihydrophysalin B (5). Hydrogenation of **2** (100 mg) in MeOH (50 cm^3) in the presence of Pt black (15 mg) under atmospheric H_2 at room temperature yielded crude dihydro derivative **5**, which was crystallized from MeOH as colorless prisms (46 mg), mp>300°C (lit,[1] >300°C).

X-Ray Crystallography. Intensity data were collected on a Rigaku AFC-5RU diffractometer equipped with a rotating anode, and using graphite-monochromated Cu $K\alpha$ (λ=1.54178 Å). Crystallographic data and experimental details are given in Table 1. The stuctures were solved with MULTAN 88 (SAYTAN),[7] and refined by full-matrix least-squares methods based upon F with $w=[\sigma(F)+(0.023F)^2]^{-1}$. Non-hydrogen atoms were refined anisotropically. Hydrogen atoms were located in a difference Fourier map and were included in the structure-factor calculation. Atomic scattering factors used were obtained from Ref. 8. All computations were performed on a FACOM M780 in the Data Processing Center of Kyoto University, using the program KPPXRAY.[9]

Results and Discussion

The perspective view of the molecular structures of

Table 3. Bond Lengths (Å) and Valence Angles (°) in **4** and **5**

Bond	**4**	**5**	Angle	**4**	**5**	Angle	**4**	**5**
C(1)–C(2)	1.475(5)	1.493(5)	C(2)–C(1)–C(10)	116.4(3)	118.1(3)	C(14)–C(15)–O(15)	125.5(3)	125.7(3)
C(1)–C(10)	1.530(5)	1.550(4)	C(2)–C(1)–O(1)	120.0(3)	120.1(3)	C(16)–C(15)–O(15)	127.4(3)	127.6(3)
C(1)–O(1)	1.223(4)	1.214(4)	C(10)–C(1)–O(1)	123.5(3)	121.7(3)	C(15)–C(16)–C(17)	99.4(2)	99.4(2)
C(2)–C(3)	1.339(6)	1.526(6)	C(1)–C(2)–C(3)	120.7(4)	108.7(3)	C(15)–C(16)–C(24)	110.1(3)	110.2(2)
C(3)–C(4)	1.480(6)	1.508(5)	C(2)–C(3)–C(4)	123.9(4)	112.0(3)	C(17)–C(16)–C(24)	114.6(3)	115.1(3)
C(4)–C(5)	1.534(5)	1.538(5)	C(3)–C(4)–C(5)	111.0(3)	111.7(3)	C(13)–C(17)–C(16)	109.3(3)	109.0(3)
C(5)–C(6)	1.449(6)	1.313(6)	C(4)–C(5)–C(6)	119.4(3)	121.6(3)	C(13)–C(17)–C(20)	102.1(3)	102.5(2)
C(5)–C(10)	1.508(5)	1.516(5)	C(4)–C(5)–C(10)	115.3(4)	115.3(3)	C(13)–C(17)–O(17)	115.9(2)	115.8(2)
C(5)–O(5)	1.445(6)		C(4)–C(5)–O(5)	112.5(3)		C(16)–C(17)–C(20)	113.7(3)	113.2(2)
C(6)–C(7)	1.496(6)	1.491(5)	C(6)–C(5)–C(10)	122.0(3)	123.0(3)	C(16)–C(17)–O(17)	104.4(2)	104.5(2)
C(6)–O(5)	1.446(5)		C(6)–C(5)–O(5)	59.9(3)		C(20)–C(17)–O(17)	111.8(3)	112.1(3)
C(7)–C(8)	1.543(6)	1.528(6)	C(10)–C(5)–O(5)	114.1(3)		C(13)–C(18)–O(18)	127.4(4)	127.9(4)
C(8)–C(9)	1.552(5)	1.535(4)	C(5)–C(6)–C(7)	122.0(4)	124.9(4)	C(13)–C(18)–O(20)	109.9(3)	110.1(3)
C(8)–C(14)	1.545(5)	1.531(4)	C(5)–C(6)–O(5)	59.9(3)		O(18)–C(18)–O(20)	122.6(3)	122.0(3)
C(9)–C(10)	1.554(4)	1.567(4)	C(7)–C(6)–O(5)	115.1(3)		C(17)–C(20)–C(21)	115.1(3)	115.3(3)
C(9)–C(11)	1.554(5)	1.546(5)	C(6)–C(7)–C(8)	117.1(4)	114.0(4)	C(17)–C(20)–C(22)	114.2(3)	114.7(3)
C(10)–C(19)	1.535(5)	1.556(5)	C(7)–C(8)–C(9)	113.9(3)	111.6(3)	C(17)–C(20)–O(20)	101.2(3)	101.1(3)
C(11)–C(12)	1.551(4)	1.547(4)	C(7)–C(8)–C(14)	108.5(3)	107.9(3)	C(21)–C(20)–C(22)	110.3(4)	110.4(3)
C(12)–C(13)	1.509(6)	1.516(5)	C(9)–C(8)–C(14)	110.5(3)	110.7(3)	C(21)–C(20)–O(20)	109.9(3)	109.7(3)
C(13)–C(17)	1.561(5)	1.556(5)	C(8)–C(9)–C(10)	113.9(3)	112.6(3)	C(22)–C(20)–O(20)	105.3(3)	104.6(2)
C(13)–C(18)	1.542(4)	1.537(4)	C(8)–C(9)–C(11)	111.2(3)	114.9(2)	C(20)–C(22)–C(23)	114.6(4)	114.4(4)
C(13)–O(13)	1.419(4)	1.420(4)	C(10)–C(9)–C(11)	109.3(3)	109.0(3)	C(20)–C(22)–O(22)	108.1(3)	107.1(3)
C(14)–C(15)	1.526(5)	1.531(5)	C(1)–C(10)–C(5)	104.1(3)	106.3(3)	C(23)–C(22)–O(22)	111.1(3)	111.2(3)
C(14)–O(14)	1.388(4)	1.407(4)	C(1)–C(10)–C(9)	108.4(2)	108.3(2)	C(22)–C(23)–C(24)	109.8(3)	110.4(3)
C(15)–O(17)	1.446(4)	1.450(4)	C(1)–C(10)–C(19)	110.3(3)	109.7(3)	C(16)–C(24)–C(23)	106.4(3)	105.7(2)
C(15)–C(16)	1.507(4)	1.516(4)	C(5)–C(10)–C(9)	111.5(3)	112.3(3)	C(16)–C(24)–C(25)	114.5(3)	114.3(2)
C(15)–O(15)	1.209(5)	1.202(4)	C(5)–C(10)–C(19)	108.8(3)	107.9(3)	C(16)–C(24)–C(28)	108.1(3)	108.2(3)
C(16)–C(17)	1.546(4)	1.554(4)	C(9)–C(10)–C(19)	113.3(3)	112.1(3)	C(23)–C(24)–C(25)	107.5(3)	108.1(3)
C(16)–C(24)	1.559(5)	1.568(4)	C(9)–C(11)–C(12)	120.3(3)	121.5(3)	C(23)–C(24)–C(28)	110.1(3)	111.1(3)
C(17)–C(20)	1.544(5)	1.545(5)	C(11)–C(12)–C(13)	118.1(3)	118.5(3)	C(25)–C(24)–C(28)	110.1(3)	109.4(3)
C(17)–O(17)	1.429(4)	1.429(4)	C(12)–C(13)–C(17)	118.3(3)	118.9(3)	C(24)–C(25)–C(26)	117.9(3)	117.3(3)
C(18)–O(18)	1.196(6)	1.193(5)	C(12)–C(13)–C(18)	112.2(4)	112.3(3)	C(24)–C(25)–C(27)	117.1(4)	116.9(3)
C(18)–O(20)	1.351(6)	1.364(5)	C(12)–C(13)–O(13)	111.4(4)	111.1(2)	C(26)–C(25)–C(27)	110.3(3)	111.4(3)
C(20)–C(21)	1.534(6)	1.536(5)	C(17)–C(13)–C(18)	97.0(3)	97.5(3)	C(25)–C(26)–O(22)	119.2(3)	119.9(3)
C(20)–C(22)	1.536(6)	1.541(5)	C(17)–C(13)–O(13)	112.0(4)	110.9(3)	C(25)–C(26)–O(26)	123.3(4)	123.0(3)
C(20)–O(20)	1.463(5)	1.465(4)	C(18)–C(13)–O(13)	104.2(3)	104.5(2)	O(22)–C(26)–O(26)	117.1(4)	116.9(3)
C(22)–C(23)	1.490(6)	1.491(6)	C(8)–C(14)–C(15)	116.5(3)	117.6(3)	C(25)–C(27)–O(14)	117.2(3)	116.8(2)
C(22)–O(22)	1.453(5)	1.459(5)	C(8)–C(14)–O(14)	107.7(3)	107.2(2)	C(5)–O(5)–C(6)	60.2(3)	
C(23)–C(24)	1.521(5)	1.521(5)	C(8)–C(14)–O(17)	110.3(2)	110.5(2)	C(14)–O(14)–C(27)	118.6(3)	118.5(2)
C(24)–C(25)	1.540(6)	1.550(5)	C(15)–C(14)–O(14)	109.2(2)	108.8(2)	C(14)–O(17)–C(17)	110.1(2)	110.1(2)
C(24)–C(28)	1.519(6)	1.512(5)	C(15)–C(14)–O(17)	104.2(2)	104.5(2)	C(18)–O(20)–C(20)	111.3(3)	111.0(2)
C(25)–C(26)	1.511(6)	1.510(5)	O(14)–C(14)–O(17)	108.7(3)	107.9(3)	C(22)–O(22)–C(26)	120.6(3)	120.8(3)
C(25)–C(27)	1.540(6)	1.532(4)	O(14)–C(15)–O(16)	107.0(3)	106.6(3)			
C(26)–O(22)	1.353(5)	1.364(4)						
C(26)–O(26)	1.195(5)	1.202(5)						
C(27)–O(14)	1.438(5)	1.457(4)						

4 and **5** in the crystal are shown in Fig. 1 and the final atomic coordinates are given in Table 2. The stereostructures of the two compounds are very similar to each other as seen from Fig. 1. The bond lengths and valence angles are summarized in Table 3, which also indicates the close similarity of **4** and **5**, i.e., the corresponding bond lengths agree within 0.02 Å and angles within 1.1° except those in the A and B ring moiety. Both crystals belong to the same space group $P2_1$ and possess similar lattice constants, and the corresponding atomic coordinates are also similar, indicating similar molecular packings of **4** and **5** in the crystals. Large sp^3 valence angles were observed at C(25), i.e., ∠C(24)–C(25)–C(26) 118° in **4** and 117° in **5** and ∠C(24)–C(25)–C(27) 117° in **4** and **5**. The unusually facile hydroxylation at this position using activated carbon catalyst[10] might be related to this partial planarity which easily generates an sp^2-like intermediate or transition state.

The only structural difference between **3** and **5**, other than the absence of 7α-OH group in **5**, is the presence of a σ-bond between C(27) and O(14) which constitutes an additional seven-membered ring in **5**. The whole molecular shape of the octacyclic **5**, with the C(27)–O(14) linkage, is similar to that of the heptacyclic **3**,

Table 4. ^{1}H NMR Spectral Data of Physalin B (**2**), 5α,6α-Epoxyphysalin B (**4**), and 2,3-Dihydrophysalin B (**5**) in DMSO-d_6 Solution (chemical shift δ, peak multiplicity, and coupling constant in Hz)

Assignments	**2**	**4**	**5**
2	5.80 dd ($J_{2,3}$=10) ($J_{2,4\beta}$=2)	5.84 dd ($J_{2,3}$=10) ($J_{2,4\beta}$=2.5)	2.27 m (α)
			2.47 m (β)
3	6.89 ddd ($J_{3,2}$=10) ($J_{3,4\alpha}$=5) ($J_{3,4\beta}$=2)	6.82 ddd ($J_{3,2}$=10) ($J_{3,4\alpha}$=5) ($J_{3,4\beta}$=2.5)	1.59 m (α)
			1.85 m (β)
4	2.89 dd (α) ($J_{4\alpha,4\beta}$=20) ($J_{4\alpha,3}$=5)	1.79 dd (α) ($J_{4\alpha,4\beta}$=20) ($J_{4\alpha,3}$=5)	ca. 2.1 m (α)
	3.27 br d (β) ($J_{4\beta,4\alpha}$=20)	3.15 dt (β) ($J_{4\beta,4\alpha}$=20) ($J_{4\beta,3}$=$J_{4\beta,2}$=2.5)	ca. 2.3 m (β)
6	5.59 br d ($J_{6,7\beta}$=6)	3.13 d ($J_{6,7\beta}$=5)	5.49 br d ($J_{6,7\beta}$=5)
7	ca. 2.0 m (α)	1.74 dd (α) ($J_{7\alpha,7\beta}$=16) ($J_{7\alpha,8}$=11)	2.00 m (α)
	2.21 m (β)	2.13 dt (β) ($J_{7\beta,7\alpha}$=16) ($J_{7\beta,6}$=$J_{7\beta,8}$=6)	2.16 m (β)
8	1.92 m	1.93 td ($J_{8,7\alpha}$=$J_{8,9}$=11) ($J_{8,7\beta}$=5)	ca. 2.0 m
9	2.95 dd ($J_{9,8}$=11) ($J_{9,11\beta}$=9)	2.96 dd ($J_{9,8}$=11) ($J_{9,11\beta}$=9)	2.91 br t ($J_{9,8}$=$J_{9,11\beta}$=10)
11	2.18 m (α)	1.93 m (α)	1.65 m (α)
	ca. 1.1 m (β)	0.98 m (β)	ca. 1.0 m (β)
12	2.17 m (α)	2.11 m (α)	1.97 m (α)
	1.45 m (β)	1.42 br dd (β) ($J_{12\beta,12\alpha}$=16) ($J_{12\beta,11\beta}$=10)	1.40 br dd (β) ($J_{12\beta,12\alpha}$=16) ($J_{12\beta,11\beta}$=10)
13	6.28 s (OH)	6.24 s (OH)	6.37 s (OH)
16	2.86 s	2.81 s	2.85 s
19	1.09 s (CH_3)	1.19 s (CH_3)	1.05 s (CH_3)
21	1.78 s (CH_3)	1.80 s (CH_3)	1.79 s (CH_3)
22	4.56 dd ($J_{22,23R}$=3) ($J_{22,23S}$=2)	4.55 m	4.57 dd ($J_{22,23R}$=3) ($J_{22,23S}$=2)
23[a]	2.14 m (R)	2.10 br d (R) ($J_{23R,23S}$=12)	2.11 dd (R) ($J_{23R,23S}$=15) ($J_{23R,22}$=3)
	1.96 m (S)	1.93 br d (S) ($J_{23S,23R}$=12)	1.92 dd (S) ($J_{23S,23R}$=15) ($J_{23S,22}$=2)
25	2.88 br d ($J_{25,27S}$=4)	2.86 d ($J_{25,27S}$=4)	2.89 d ($J_{25,27S}$=4)
27	3.60 dd (R)[b] ($J_{27R,27S}$=14) ($J_{27R,25}$=1)	3.56 br d (R) ($J_{27R,27S}$=14)	3.60 d (R) ($J_{27R,27S}$=13)
	4.26 dd (S)[b] ($J_{27S,27R}$=14) ($J_{27S,25}$=4)	4.24 dd (S) ($J_{27S,27R}$=14) ($J_{27S,25}$=4)	4.25 dd (S) ($J_{27S,27R}$=13) ($J_{27S,25}$=4)
28	1.16 s (CH_3)	1.15 s (CH_3)	1.15 s (CH_3)

a) Configurational assignment is based on the observation of NOESY cross peak between H-16 and H-23*S*. b) Configurational assignment in Ref. 2 should be revised.

in which the C(27)–methyl is in van der Waals' contact with the C(14)–OH group [C(27)···O(14) distance 3.34 Å]. Some corresponding torsion angles in **3** and **5**, however, are significantly different from each other [∠C(9)–C(11)–C(12)–C(13) −43° in **3** and 0° in **5**; ∠C(14)–C(15)–C(16)–C(24) −120° in **3** and −89° in **5**].

The ^{1}H NMR characteristics of physalin B (**2**) and related physalins possessing the C(14)–O–C(27) bridge (referred to as type B), compared to those of physalin A-type physalins and their derivatives which lack the acetalic linkage (type A), can be explained in terms of these structural differences. The ^{1}H NMR spectra of **2**, **4**, and **5** measured in DMSO-d_6 solutions are summarized in Table 4.

The 12β-proton in all these physalins belonging to type B resonates at δ=1.40—1.45, i.e., at much higher field than that in type A (δ=ca. 1.9).[2,4] This is reasonably explained by the torsion angle difference around the C(11)–C(12) bond described above which causes the larger anisotropic shielding effect of the C(15)–carbonyl in the former compounds. Another torsion angle [∠C(14)–C(15)–C(16)–C(24)] which shows large difference between **3** and **5** can be related to the chemical shift of the C(28)–methyl group. The methyl singlet in type B physalins invariably appears at δ=1.15—1.20,[2] while in type A it resonates at lower field (**1** δ=1.55, **3** δ=1.47, etc.) in most cases.[2,4,11] The ring closure from type A to type B brings about the locational change of the C(28)–methyl group from the anisotropic deshielding to the shielding region of the C(15)–carbonyl group. The shielding effect of the C(15)–carbonyl is most pronounced on the 11β-H (δ=ca. 1.0) for both type A[4] and type B physalins. The C(27)–methylene of type B physalins also exhibited large chemical shift difference between the diastereotopic protons. The *pro-R* hydrogen, which shows a cross peak with the C(28)–methyl

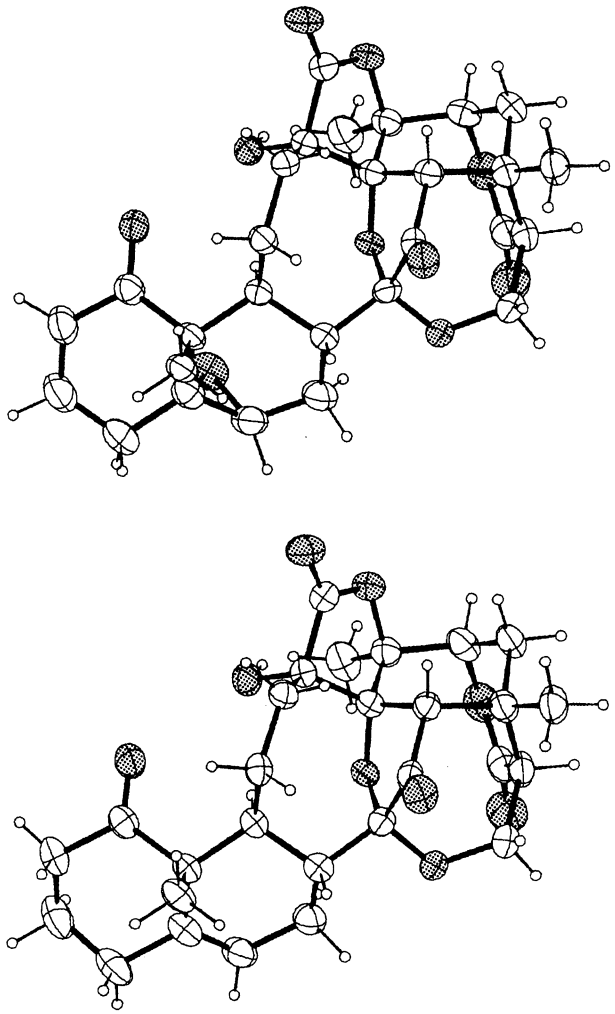

Fig. 1. Perspective view of the molecules of 5α,6α-epoxyphysalin B (**4**, upper) and 2,3-dihydrophysalin B (**5**, lower).

in NOESY spectra, is observed at higher field compared to its *pro-S* counterpart. The smaller and larger vicinal coupling constants $J_{27,25}$ of the *pro-R* (ca. 0) and *pro-S* (ca. 4 Hz) proton signals are in agreement with the torsion angles ∠H–C(25)–C(27)–H, ca. +67° and ca. −51°, respectively.[12] In compound **4** the allylic C(4)–methylene protons adjacent to the epoxy group exhibits quite a large chemical shift differene, which is consistent with the reported spectral data of a withanolide possessing the same partial structure as **4**.[13]

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