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MINOR DITERPENOIDS FROM *PORTULACA* CV JEWEL

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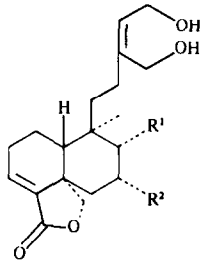
Key Word Index—*Portulaca* cv Jewel; Portulacaceae; diterpene; *trans*-clerodane; bicyclo[5.4.0]undecane skeleton, jewenol A and B; chemotaxonomy.

Abstract—A *trans*-clerodane and a diterpenoid with the bicyclo[5.4.0]undecane skeleton were isolated from the aerial part of *Portulaca* cv Jewel, and their structures were elucidated by spectroscopic methods, chemical correlation and X-ray diffraction analysis. The chemotaxonomical significance is discussed.

INTRODUCTION

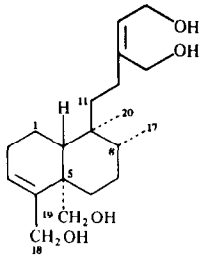
In view of the biosynthetic and chemotaxonomical interest we have been studying the diterpenoid constituents of *Portulaca* plants [1–5]. In our previous paper, we reported the isolation of *trans*-clerodane type diterpenoids **1–4** as major products from *P.* cv Jewel [3]. An investigation on the minor constituents of the same plant has demonstrated the occurrence of a diterpenoid of a different type, in addition to a new clerodane compound.



1 R^1 = CH_2OH, R^2 = H
2 R^1 = Me, R^2 = H
3 R^1 = Me, R^2 = OH
4 R^1 = CHO, R^2 = H

RESULTS AND DISCUSSION

The chromatographic fractions of an ethyl acetate extract of *P.* cv Jewel, other than those of portulide A–D, were further separated to give two new diterpenoids, designated as jewenol A (**5**) and B (**6**), respectively. The structures of both compounds were elucidated by spectroscopic methods, the former being of the clerodane type and the latter having a bicyclo[5.4.0]undecane skeleton. The structure of jewenol B (**6**) was confirmed by X-ray diffraction analysis.



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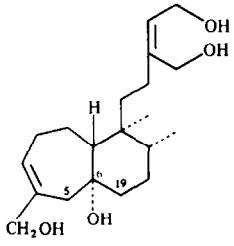

6

The first compound, jewenol A (**5**) has the molecular formula $C_{20}H_{34}O_4$ as deduced from the $[M-2H_2O]^+$ peak in the high resolution mass spectrum. The molecular ion peak was ascertained by negative ion FAB mass spectroscopy in glycerol $[M+\text{glycerol}-H]^-$. The IR spectrum of **5** indicated the presence of hydroxyl groups, and the lack of the γ-butenolide ring as observed in portulides. The inspection of ^1H and ^{13}C NMR spectra of **5** indicated the presence of 5-hydroxy-3-hydroxymethyl-3-pentenyl side chain, a characteristic for the diterpenoids found in *Portulaca* plants. The signals due to two hydroxymethylene groups were observed at δ64.47 and 65.64 (C-18 and C-19, respectively) in the ^{13}C NMR spectrum, and as two pairs of AB-type signals at δ3.81, 4.12 (H-18) and δ3.58, 3.97 (H-19), respectively, in the ^1H NMR spectrum. Furthermore, the ^1H NMR spectrum showed signals of a tertiary methyl group at δ0.78 (H-20) and a secondary methyl at δ0.86 (H-17). The olefinic proton (H-3) in the A ring was observed at δ5.71, not as double doublets as in the case of compounds **1–4**, but as a broad triplet. The chemical shift of H-3 was at the shielded position as compared with that of compounds **1–4**. These spectral data suggested that jewenol A had the clerodane structure **5**, which was consistent with the result of ^1H NMR analysis based on (homo nuclear spin decoupling HNSD) experiments and the ^{13}C NMR spectrum assigned by INEPT and ^1H-selective decoupling. The ^{13}C NMR spectrum was assigned by INEPT and ^1H-selective decoupling. This structure assignment was confirmed by conversion of portulide B (**2**) to **5** through diisobuthylaluminium hydride reduction.

The second compound, jewenol B (**6**) has the molecular formula $C_{20}H_{34}O_4$ as revealed from the M^+ peak in the high resolution mass spectrum. The IR spectrum showed the presence of hydroxyl groups. The ^1H NMR spectrum

Table 1. ^{1}H NMR spectral data of compounds **5** and **6** (400 MHz, CD_3OD, TMS as internal standard)

H	5	6
1	1.67*	1.68*, 1.40*
2	2.17*, 2.25*	α 2.28 *br dt* (13.9, 6.5)
		β 2.07*
3	5.71 *br t* (3.7)	5.99 *m*
5	—	α 2.40 *br d* (13.9)
		β 2.04 *d* (13.9)
6	1.17 *td* (12.5, 4.6)	—
	2.22*	
7	1.44*, 1.49*	1.30*, 1.55*
8	1.60*	1.57*
10	1.65*	1.47 *d* (11.7)
11	1.45*, 1.56*	1.40*, 1.53*
12	1.92 *td* (14.2, 5.0)	2.02*
	2.03 *td* (14.2, 4.6)	
14	5.46 *t* (6.6)	5.47 *t* (6.8)
15	4.12 *d* (6.6)	4.14 *d* (6.8)
16	4.09 *s*	4.10 *s*
17	0.86 *d* (6.8)	0.85 *d* (6.8)
18	3.81 *d* (12.0)	3.88 *br s*
	4.12 *d* (12.0)	
19	3.58 *d* (10.5)	1.6 ~ 1.75
	3.97 *d* (10.5)	
20	0.78 *s*	0.80 *s*

*Overlapped signal.

Table 2. ^{13}C NMR spectral data of compounds **5** and **6**. (100 MHz, CD_3OD)

C	5	6*
1	18.27	23.11
2	27.24	29.60
3	128.83	131.23
4	146.30	140.67
5	43.99	47.58
6	32.19	72.61
7	28.00	29.02
8	37.57	38.68
9	39.79	41.86
10	47.59	55.08
11	38.30	38.04
12	29.11	29.94
13	143.46	144.61
14	127.22	128.07
15	58.63	59.64
16	60.08	61.24
17	16.19	17.50
18	64.47	69.47
19	65.64	43.76
20	19.30	18.32

*Assignments of **6** were made with the aid of ^{13}C–^{1}H COSY experiments.

of jewenol B (**6**) (Table 1) indicated the presence of a 5-hydroxy-3-hydroxymethyl-3-pentenyl side chain, a tertiary methyl group (δ0.80, H-20), and a secondary methyl group (δ0.85, H-17). A pair of AB-type signals was present at δ2.04 (H_β-5) and 2.40 (H_α-5) reminiscent of the one observed in the case of portulal and portulene [4]. This fact suggested that the A ring should be 7-membered. The olefinic proton in the A ring was observed at δ5.99 as a multiplet. Furthermore, the signal due to a hydroxy methylene attached to the olefinic bond appeared at δ3.88 as a broad singlet. These data, in cojunction with the ^{13}C NMR spectrum (Table 2), led to the assumption of a bicyclo[5.4.0]undecane structure like portulene [4] for jewenol B. The tertiary hydroxyl group corresponding to a carbon signal at δ72.61 (*s*) was located to C-6, since the signals of C-5 and C-19 carbons was deshielded somewhat as compared with portulene. Thus the structure **6** was assigned for jewenol B. This assignment was verified by single crystal X-ray analysis.

Crystals of jewenol B (**6**) were grown from methanol–water solution. The crystals were orthorhombic and belong to chiral space group $P2_1$. Accurate cell constants determined from a least-squares fit of 15 high angle reflections were: $a = 12.153(7)$, $b = 7.943(5)$, $c = 10.242(8)$Å and $\beta = 99.17(6)°$. All unique diffraction maxima with $2\theta < 55°$ were recorded in the $\omega/2\theta$ scan mode using a four circle diffractometer and graphite monochromated MoK_α X-rays. Three reference reflections monitored every 100 reflections displayed neither systematic nor significant deviations from their initial intensities. Of the 2422 reflections surveyed, 1870 (77%) were judged observed [$I > 3\sigma(I)$] after correction for Lorentz, polarization and background effects. The structure was solved by MULTAN [6]. All hydrogen atoms were located on a difference electron density synthesis and included in subsequent calculations. Full matrix least-squares refinement with anisotropic temperture factors for the non-hydrogen atoms and isotropic temperature factor for hydrogen converged to a conventional crystallographic discrepancy index of 0.042. [7] The absolute configuration was not determined by the X-ray experiment. Additional crystallographic details such as bond distances, bond angles and, observed and calculated structure factors are available as the supplementary material.

Figure 1 gives a perspective drawing of **6** with the atomic numbering [8]. The cycloheptene ring is in a chair form. In general, bond distances and angles agree well with generally accepted values. There are four O–H---O intermolecular hydrogen bonds. All other intermolecular distances correspond to van der Waals contacts.

Present work has demonstrated that *P.* cv Jewel contains, in addition to *trans*-clerodanes, the diterpenoid with a bicyclo[5.4.0] undecane skeleton as a minor constituent, which occurs also in minor amount in *P. grandiflora* Hook. and represents the major component in *P. pilosa* L. From the biosynthetic view [4, 5] the bicyclo[5.4.0]undecane compounds arise on the way from the *ent*-labdadienyl cation to portulal (**1**) and the route leading to the clerodane compounds represents another branched pathway from the labdane intermediate. Therefore, our result indicate that *P.* cv Jewel is a relatively unique species so far as the diterpenoid constituents are concerned, but it has still retained some relationships with the other two plants. Interestingly this chemotaxonomical assumption reflects exactly the delicate situation of *P.* cv Jewel in plant systematics which has been suggested from the study of chromosome number

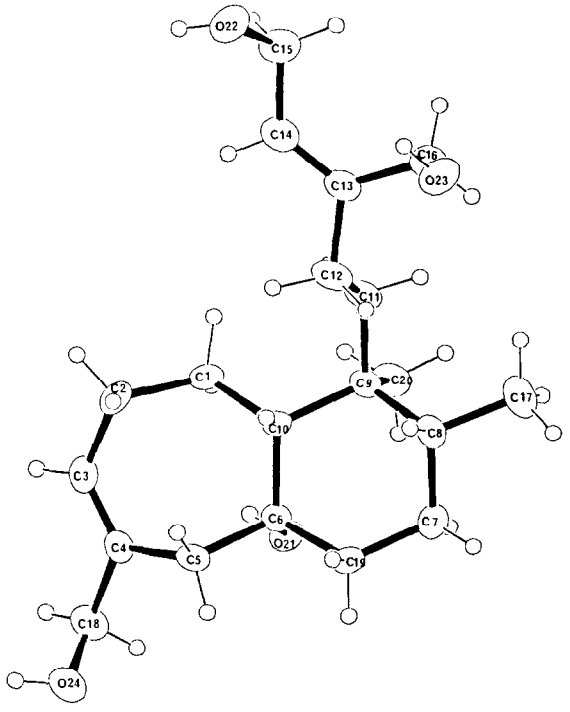

Fig. 1.

and the other genetic and morphological investigations [9].

EXPERIMENTAL

^{1}H NMR spectra were measured at 400 MHz with TMS as internal standard and ^{13}C NMR spectra were measured at 100 MHz (Jeol JNM GX-400). EIMS were taken at an ionization voltage at 70 or 22 eV (Jeol JMS HX-100). TLC was performed on silica gel plates (Merck, Kiesel gel 60, F_{254}) etc.

Plant materials. The seeds of *Portulaca* cv Jewel were purchased from the Sakata seed company (Tokyo, Japan), plants were grown at Plant Garden, Faculty of Science, Osaka City University and collected at the end of August 1983.

Extraction and isolation of diterpenoids. The aerial parts of fresh plants (27 kg) were ground with MeOH and kept at room temp. for several weeks. After filtration, the filtrate was evapd to one-tenth of original vol. treated with hexane and extracted with EtOAc. The residue (120 g) after evapn of the solvent was chromatographed on silica gel. Elution with $CHCl_3$–MeOH (19:1–9:1) gave fractions containing diterpenoids and these were separated by repeated chromatography (Merck silica gel of 230–400 mesh, RP-8 and prep. TLC) affording the four diterpenes, portulide A–D. The fractions near those containing portulide B were separated by repeated chromatography (silica gel of 230–400 mesh, K60, Merck; ODS Fujigel packed column RQ-2; prep TLC, silica gel K60, F_{254}, 0.5 mm, Merck) affording the two diterpenoids, jewenol A (20 mg) and B (44 mg).

Jewenol A. Colourless oil; $[\alpha]_D^{31} = -87.6°$ (EtOH; *c* 0.37); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3400; HRMS (DI) *m/z* 302.2247 $[M-2H_2O]^+$, calc. for $C_{20}H_{30}O_2$; negative ion FABMS (glycerol) *m/z* 429 $[M+\text{glycerol}-H]^-$

Jewenol B. mp. 123.5–124.5° (MeOH–H_2O); $[\alpha]_D^{31} -59.2°$ (EtOH; *c* 0.52); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3400; HRMS (DI) *m/z* 338.2433 $[M]^+$ calc for $C_{20}H_{34}O_4$; FABMS (glycerol) 339 $[MH]^+$ Reduction of **5**. To a THF soln (5 ml) of **5** (86 mg) was added DIBAL-H (toluene, 1.5 M soln, 1.5 ml) at 0°. The soln was stirred for 15 min at room temp. After usual work-up, the crude product was chromatographed on silica gel prep. TLC to give **6** (61 mg).

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