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STEREOCHEMICAL STUDIES ON BORONOLIDE, AN α-PYRONE FROM *TETRADENIA BARBERAE*

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Key Word Index—*Tetradenia barberae*; Lamiaceae; α-pyrone; boronolide.

Abstract—The structure of boronolide isolated from *Tetradenia barberae* has been confirmed as 6*R*-[1*R*,2*R*,3*S*-(trisacetyloxy)-heptyl]-5,6-dihydro-2H-pyran-2-one by chemical degradation.

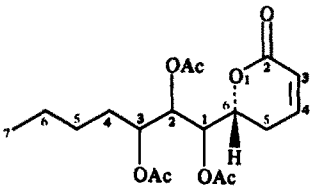
INTRODUCTION

In the course of our stereochemical studies of 6-substituted α-pyrones occurring in species of the family Lamiaceae, we have investigated the absolute configuration of C-6 in boronolide (1), isolated from *Tetradenia barberae* (N.E.Br) Codd. Boronolide has previously been obtained from *Tetradenia fruticosa* Benth. [1], a species valued in Madagascan folk medicine. The relative stereochemistry of boronolide (2) has been determined by x-ray analysis [2], and an (*R*)-configuration for C-6 proposed by application of Hudson's lactone rule [3] to the molecular rotation. In this paper we have confirmed this assignment by chemical degradation. The applicability of this method to the examination of the configuration at this chiral centre in other similar 6-substituted α-pyrones is discussed.

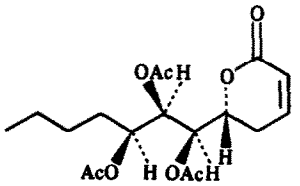
RESULTS AND DISCUSSION

The dried leaves of *T. barberae* yielded boronolide (1), $C_{18}H_{26}O_8$, mp 89–90°, $[\alpha]_D^{26}$ +28°, in 0.7% yield. The presence of an α,β-unsaturated δ-lactone was shown by the UV (λ_{max} 205 nm, log ε 3, 92) and IR (ν_{max} 1710 cm^{-1}) spectra. The chemical structure of 1 was confirmed by ^1H NMR, ^{13}C NMR and mass spectrometry. Boronolide is related to deacetylboronolide (3) and 1,2-dideacetylboronolide (4) isolated from *Tetradenia riparia* (formerly Iboza) [4] an African species widely used in central African tribal medicine [5]. The ^{13}C NMR spectra of compounds 1, 3 and 4 are compared in Table 1.

Malic acid can be derived by chemical degradation from carbon atoms 4,5,6 and 1 (side-chain) of boronolide. Hence the absolute configuration at C-6 in 1 can be related to the configuration at C-2 in malic acid. Saponification of 1 with 0.1 M KOH yielded 3, $C_{12}H_{20}O_5$, mp 99–101°, $[\alpha]_D^{22}$ +56°. Surprisingly, compound 3 has been reported as showing no optical activity [4].



1



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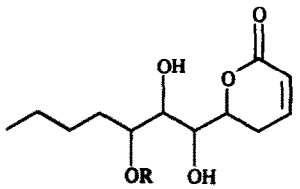
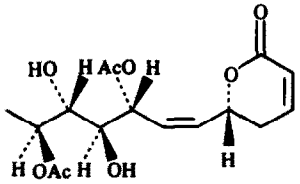


3 R = H
4 R = Ac



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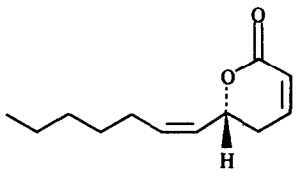
*Author to whom correspondence should be addressed.

Table 1. ^{13}CNMR spectral data of compounds 1 (75.5 MHz, $CDCl_3$) 3 (75.47 MHz, $CDCl_3$) and 4 (50.29 MHz $CDCl_3$) [4], TMS as internal standard

C	1	3	4
2	162.22	163.72	163.4
3	121.30	120.79	121.1
4	143.98	145.80	145.4
5	25.14	25.49	25.46
6	75.10	†	77.12
1 (side-chain)	70.62*	70.06*	70.22*
2 (side-chain)	70.55*	74.14*	72.07*
3 (side-chain)	71.57*	76.40*	75.56
4 (side-chian)	30.26	33.29	30.3
5 (side-chain)	27.04	27.47	27.2
6 (side-chain)	22.36	22.41	22.2
7 (side-chain)	13.86	13.77	13.74
Acetate (Me)	13.86		13.74
	20.62		20.94
	20.72		
	20.93		
Acetate (C=O)	169.45		171.6
	169.67		
	170.22		

*Assignments interchangeable.

†Signal not assigned because of overlap with solvent peaks.



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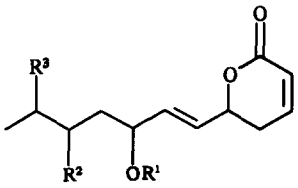
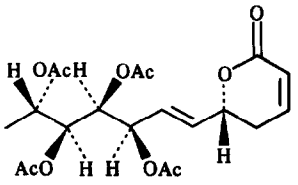


7 R^1 = Ac, R^2 = R^3 = H
8 R^1 = R^2 = R^3 = H
9 R^1 = Ac, R^2 = R^3 = OAc



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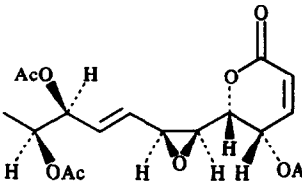
Periodate oxidation of 3 followed by oxidative ozonolysis, base hydrolysis and acidification yielded a mixture of acids. The presence of malic acid in this mixture was confirmed by GC of the acetylated dimethyl ester. Diasteromers prepared by esterification of the enantiomers of malic acid with an optically pure chiral alcohol can be separated by capillary GC on a non chiral stationary phase [6].

The absolute stereochemistry of the malic acid derived from 1 followed from conversion to the acetylated 2-(+)-dibutyl ester and GC comparison with the acetylated 2-(+)-dibutyl ester of L-malic acid and the acetylated 2-(+)-dibutyl esters of DL-malic acid; the identities of the GC peaks were confirmed by GC/MS. This method establishes an (*R*)-configuration for the derived malic acid. The main advantage of this GC method is that only relatively small amounts of the α-pyrone are required (*ca* 0.5 mmol) and extensive isolation and purification of the derived malic acid is not necessary.

Recently we established the structure and absolute stereochemistry of a similar α-pyrone synrotolide (**5**) [7], isolated from *Syncolostemon rotundifolius*. Synrotolide is related to argentilactone (**6**) [8] obtained from *Aristolochia argentina*, umuravumbolide (**7**) and deacetylumuravumbolide (**8**) [9] isolated from *Tetradenia riparia*, hyptolide (**9**) [10] from *Hyptis pectinata*, anamarine (**10**) [11] and olguine (**11**) [12] extracted from an unclassified *Hyptis species* and 5-deacetoxyepiolguine (**12**) [13] from *Hyptis oblongifolia*. With the exception of *A. argentina*, which belongs to the Aristolochiaceae, all the other species are members of the Lamiaceae.

Synrotolide has an (*R*)-configuration at C-6 [7] and this was confirmed using the degradative and GC techniques outlined here. Hence this method can be used to examine the configuration of C-6 in compounds **4**, **7**, **8** and **9**. The absolute stereochemistry of compound **11**, with an (*R*)-



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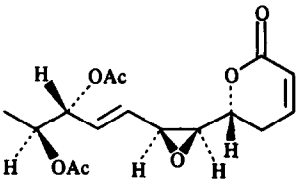

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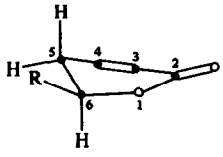

configuration at C-6, has been established by the application of statistical methods to the X-ray diffraction data [12]. This result has been used to propose the absolute stereochemistry of related compounds **10** and **12** from their X-ray data [11, 13].

Circular dichroism has been used to assign the absolute stereochemistry of compound **6** [8]. The conformation of the lactone ring of **6** shown in Fig. 1 was proposed by application of Snatzke's rules [14, 15] to the positive Cotton effect at 256 nm. The pseudo-equatorial orientation of the heptenyl side chain followed from NMR studies [8]. Compounds **1** ($\Delta\varepsilon = 2.48$ at 256 nm) and **5** ($\Delta\varepsilon = 2.45$ at 266 nm) have a positive cotton effect near 260 nm, as do compounds **10** [11] and **11** [12]. If an equatorial orientation of the side-chain (Fig. 1) is assumed in compounds **1**, **5**, **10** and **11**, then circular dichroism provides further evidence for an (*R*)-configuration of C-6 in these compounds. It is probable that compounds **4**, **7**, **8** and **9** also possess this configuration.

EXPERIMENTAL

All GC experiments were performed with a Durabond fused silica capillary column coated with DB 225 and using an FID detector. Oven temp. 165°, injector temp. 250°; detector temp. 250°, He carrier at 1.0 ml/mm, split ratio 1:50, injection volume 0.5 μl. The 70 eV mass spectra of the derivatised malic acids were recorded on an HP 5988A mass spectrometer in combination with a Durabond DB 225 fused silica capillary column. Oven temp. 160°, injector temp. 250°. *T. barberae* (Albany museum, Grahamstown Voucher No. A 7362) was collected in the Fish river valley near Grahamstown in March, 1986.

Isolation. Air-dried leaves of *T. barberae* (577 g) were soaked in Me_2CO (25 l) for 4 days. The soln was concentrated by flash distillation, decolourized with activated charcoal (BDH) and filtered through a celite pad. The remaining Me_2CO was removed *in vacuo* to give a dark brown gum (49.1 g). This gum (16 g) was chromatographed on silica gel (Merck No. 7734) (220 g) and eluted with hexane–EtOAc. The fractions eluted with hexane–EtOAc (1:1) showed a single spot on TLC. These fractions were combined and the hexane–EtOAc removed *in vacuo*. Compound **1** (1.2 g) was crystallized from EtOH. Recrystallization from hexane–C_6H_6 (5:1) yielded white needles (0.9 g); mp 89–90° (lit. 90°) [1]; $[\alpha]_D^{26}$ +28° (EtOH; *c* 0.08) (lit. +25°) [1]; UV λ_{max}^{MeOH} nm: 205 ($\log\varepsilon$ 3.92); IR ν_{max}^{KBr} cm^{-1}: 1730 (ester carbonyl), 1710 (α,β-unsatd δ-lactone) 1365, 1220, 1020, 952; EIMS (70 eV) *m/z* (rel. int.): $[M^+ + 1]$ 371 (2), 311 (3), 273 (23), 242 (47), 201 (36), 18 (47), 172 (28), 171 (39), 159 (36), 140 (55), 121 (47) 110 (42), 97 (46), 95 (40), 68 (42), 43 (100); ^{1}H NMR (300 MHz, $CDCl_3$): δ0.84 (3H, *t*, $J_{6,7} = 7$ Hz, H-7 side-chain), 1.24 (4H, *m*, H-5, H-6 side-chain), 1.53 (2H, *m*, H-4 side chain), 2.04 (1H, *s* acetyl), 2.07 (1H, *s*, acetyl), 2.11 (1H, *s*, acetyl), 2.28 (1H, *m*, H-5' ring), 2.49 (1H, *m*, H-5 ring), 4.50 (1H, *m* H-6 ring), 4.80 (1H, *dt*, H-3 side chain), 5.31 (2H, *ddd*, H-1, H-2 side chain), 5.95 (1H, *m*, H-3 ring), 6.84 (1H, *m*, H-4 ring), CD (MeOH), 306 nm ($[\theta] = 0$), max 256 ($[\theta] = +8188$). (Found: C, 58.01; H, 7.22.. $C_{18}H_{26}O_8$ requires C, 58.37; H, 7.08%).

Saponification of compound **1**. Saponification of **1** with 0.1 M KOH at room temp. gave **3**, mp 99–100° (2:1 hexane–C_6H_6) (lit. 99–100°) [1]; $[\alpha]_D^{22}$ +56° (EtOH; *c* 0.07) (lit+48°) [1]; IR ν_{max}^{KBr} cm^{-1}: 3240 (OH), 1695 (α,β-unsatd δ-lactone) 1440, 1244, 1062, 1015, 970.

Periodate oxidation and ozonolysis of compound **3**. $NaIO_4$ (0.13 mmol) was stirred with an aq. soln of **3** (0.61 mmol) for 16 hr at room temp. The soln was freeze-dried and the pale yellow residue extracted with EtOAc (3 × 100 ml). Removal of the EtOAc *in vacuo* gave 92 mg of a yellow oil, which was dissolved in 20 ml of HOAc–HCOOH (9:1). Excess ozone was passed through this solution at 10°. The soln was purged with N_2 after which it was refluxed at 110° for 12 hr, while a stream of O_2 was passed through. Evaporation of the solvent left a dark yellow residue which was stirred with 4% KOH (10 ml) for 30 min. The basic soln was then passed through an IE resin (Amberlite IR-120). The acidic eluate was freeze-dried to afford a brown oil (70 mg). The presence of malic acid in a portion of this oil (20 mg) was confirmed by esterifying the oil with excess ethereal–CH_2N_2, followed by treatment with Ac_2O–pyridine at 100° for 30 mins. The products were compared by GC (oven temp. 150°) with the same derivative of L-malic acid. An R_t of 11.23 min was obtained for both compounds.

Oxidative ozonolysis of compound **5**. An excess of ozone was passed through a solution of **5** [7] (0.30 mmol) in 20 ml of HOAc–HCOOH (9:1) at 10°. Oxidative work-up of the soln followed by base hydrolysis (4% KOH), acidification by filtration through an IE resin and final freeze-drying gave a brown oil (41 mg).

The absolute stereochemistry of the malic acid derived from compounds **1** *and* **5**. The acetylated 2-(+)-dibutyl ester of 10 mg of DL malic acid was prepared [6]. GC under the conditions stated gave two peaks R_t 17.90 and 18.26 min, of equal area, corresponding to the two diastereomers L-(++) and D-(++). GC of the acetylated 2-(+)-dibutyl ester of L-malic acid (10 mg) similarly prepared, showed that the second peak corresponded to the L-(++)-diastereomer. Preparation of the same derivative of the oxidative ozonolysis products of compounds **1** (40 mg) and **5** (41 mg) gave a major peak at R_t 17.85 min and R_t 17.90 min, respectively, thus establishing a D-configuration for the malic acid derived from these compounds. The identity of the peaks was confirmed using GMS. The mass spectrum of the acetylated 2-(+)-dibutyl ester of D-malic acid derived from **1** had *m/z* (rel. int.): No $[M]^+$, 215 (5) $[M - OC_4H_9]$, 188 (3), 177 (5), 159 (100) $[M - OC_4H_9 - C_4H_8]$, 132 (31), 131 (22), 99 (18), 89 (49), 71 (33), 57 (31) C_4H_9.

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