Pergamon

Tetrahedron 58 (2002) 3065–3071

TETRAHEDRON

Structure, conformation and absolute configuration of novel bisnorsesquiterpenes from the *Adesmia boronioides* essential oil

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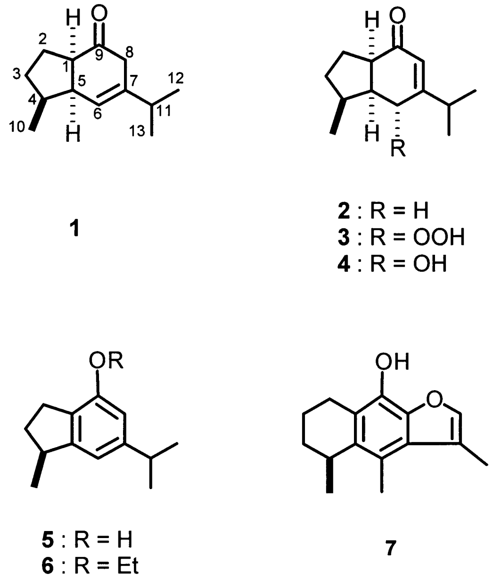
Received 16 January 2002; revised 14 February 2002; accepted 18 February 2002

Abstract—Two novel bisnorsesquiterpenes (1*R*,4*S*,5*R*)-esquel-6-en-9-one (**1**) and (1*R*,4*S*,5*S*)-esquel-7-en-9-one (**2**) were isolated from the essential oil of *Adesmia boronioides*. Their structures and relative stereochemistry were elucidated by 1D and 2D NMR data, including carbon–carbon connectivity and NOESY experiments. The absolute configuration was determined by chemical conversion of **1** into phenol **6**, whose Cotton effect was compared with that of cacalol. The conformational behavior of the new compounds containing the hydrindane skeleton was studied by molecular mechanics. Compound **1** possesses a very pleasant fruity odor. © 2002 Elsevier Science Ltd. All rights reserved.

1. Introduction

Adesmia boronioides is an aromatic and medicinal plant, which grows profusely in the Patagonian region of Argentina, from Neuquén to the island of Tierra del Fuego, and also in southern Chile.[1] A noticeable characteristic of this member of the Leguminosae family is the presence of a large number of vesicles in the leaves,[2] which produce important amounts of oil. This species has been used by local people for the treatment of rheumatic pains, hair loss,[3] colds, digestive disorders,[4] as well as an aphrodisiac.[5] Only a few studies on the chemical composition of other members of this large genus have been carried out.[6,7] During our investigations on the chemical composition of Patagonian aromatic plants, we detected that the essential oil of the aerial parts of this plant exhibited a very pleasant fruity odor, which could be interesting to the perfume industry. This fact prompted us to undertake a detailed chemical study, which yielded two novel bisnorsesquiterpenes. Although substances containing 13 carbon atoms are rare in nature, several of them are widely used as fragrant ingredients.[8]

Since the compounds obtained in the present study are new bisnorsesquiterpenes (Fig. 1), we named them (1*R*,4*S*,5*R*)-esquel-6-en-9-one (**1**) and (1*R*,4*S*,5*S*)-esquel-7-en-9-one (**2**) to remember their origin in Esquel, Argentina. Herein, we report the stereostructure, absolute configuration as well as a conformational analysis based on molecular mechanics studies.[9]



1

2 : R = H
3 : R = OOH
4 : R = OH

5 : R = H
6 : R = Et

7

Figure 1. Structure and absolute configuration of esquelane derivatives **1**–**4**, indane derivatives **5** and **6**, and cacalol (**7**).

Keywords: bisnorsesquiterpenes; stereochemistry; conformation; molecular mechanics; absolute configuration; *Adesmia boronioides*.

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PII: S0040-4020(02)00191-6

2. Results and discussion

(1*R*,4*S*,5*R*)-Esquel-6-en-9-one (**1**) displayed a molecular ion $[M]^+$ at *m/z* 192.1510 by HREIMS, in agreement with the molecular formula $C_{13}H_{20}O$. A strong IR absorption at 1702 cm^{-1} indicated the presence of a six-membered ring ketone, while the ^1H and ^{13}C NMR spectra confirmed the bisnorsesquiterpene-type structure. The ^1H NMR spectrum showed signals for a vinylic hydrogen, a methylene α to a carbonyl group, an isopropyl moiety and a secondary methyl group. The DEPT spectrum indicated the presence of three methyl groups, three methylenes, five methines and two quaternary carbon atoms. The low-field signals revealed the presence of a carbonyl group at δ 212.2 and a trisubstituted double bond at δ 141.3 and 118.3. A ^{13}C–^{13}C connectivity experiment carried out using the CCC2D pulse sequence allowed us to secure the whole hydrocarbon framework of structure **1**. Furthermore, a ^1H–^{13}C HSQC experiment was very useful to assign the sp^3 methylene and methine region, where three pairs of methylene and four methine proton signals were recognized. It is relevant to mention that the coupling constant between the two hydrogen atoms attached to the ring fusion was indicative of a *cis*-fused hydrindane system ($J_{1,5}$=9.0 Hz), as it has been observed in other molecules containing this ring system.[10]

(1*R*,4*S*,5*R*)-Esquel-7-en-9-one (**2**) showed a molecular ion $[M]^+$ at *m/z* 192.1507 by HREIMS, also in agreement with the molecular formula $C_{13}H_{20}O$. The IR absorption at 1650 cm^{-1} suggested the presence of an α,β-unsaturated six-membered ring ketone, which was confirmed by the UV absorption at 232 nm. On the other hand, the ^{13}C NMR spectrum evidenced the bisnorsesquiterpene skeleton, revealing that the only difference with respect to compound **1** was the position of the double bond. The signals for the α,β-unsaturated ketone were found at δ 202.6, 169.7 and 122.4. HSQC and HMBC experiments allowed us to verify the structure of **2** as well as the ^1H and ^{13}C NMR assignments. In this case, the coupling constant for the ring fusion hydrogens was $J_{1,5}$=6.4 Hz. Additionally, compound **1** was isomerized into **2** by treatment with $NaHCO_3$ in MeOH/H_2O.

Compound **1** underwent oxidation upon exposition to air for several weeks to give hydroperoxide **3**, even when stored at −20°C. The reaction time was substantially decreased by stirring a solution of **1** in pentane under an O_2 atmosphere. Furthermore, treatment of hydroperoxide **3** with KI afforded alcohol **4**. The 500 MHz ^1H NMR spectrum of hydroperoxide **3** displayed a good dispersion of the proton resonance signals, which in turn allowed us to clearly observe and assign the multiplicities and coupling constants for most of the proton signals. Molecular modeling of **3** in combination with a fully assigned NOESY spectrum allowed us to establish the relative stereochemistry of this substance. A molecular mechanics study of **3** was conducted using a systematic search procedure[11] with the aid of Dreiding models to select the range of torsion angle movements and employing the MMX force field[9] as implemented in the PCModel program. The global minimum for peroxide **3** was found at 20.56 kcal/mol (**3c**, Table 1) having a slightly distorted twist conformation for the five-membered ring and a distorted envelope conformation for the cyclohexenone moiety. Molecular dynamics calculations were performed over the minimum energy structure using the leap-frog Verlet algorithm[12] as implemented in the Dynam routine. According to these calculations, twelve minimum energy conformations were detected for compound **3** (Table 1), which mainly arose from the pseudorotation circuit of the five-membered ring and rotation of the isopropyl moiety. Four representative conformations for this substance are depicted in Fig. 2. In these structures (**3a**, **3d**, **3h** and **3j**) the H(1)–C(1)–C(5)–H(5) dihedral angles are −44, −35, +12 and +41°, respectively. Structures **3a**, **3d**, **3h** and **3j** fully explain the NOESY correlations between H-1/H-4, H-1/H-5, H-3α/H-4, H-3α/H-5, H-3β/Me-10, H-4/H-5, which allowed us to determine the stereochemistry at C-1, C-4 and C-5 of **3**, and therefore, that for esquelane derivatives **1**, **2** and **4**. Additionally, NOESY correlations between H-3β/H-6, H-4/H-6 and H-6/Me-10 were in full agreement with the α-orientation of the peroxide moiety at C-6 in **3**, and also with the α-orientation of the hydroxyl group in compound **4**.

Additionally, the molecular mechanics/dynamics study was extended to esquelane derivatives **1**, **2** and **4** using the same protocol, which allowed us to estimate the flexibility and the conformational trajectory present in esquelane derivatives. For compound **1**, a series of fifteen minimum energy structures were found with the global minimum at 18.17 kcal/mol at a H(1)–C(1)–C(5)–H(5) dihedral angle of −40°. In contrast, the molecular dynamics study of **2** indicated that this substance is less flexible than **1**. Using the same parameters than those for **1** and **3**, compound **2** yielded only three conformations at 21.44, 21.80 and 21.94 kcal/mol with a H(1)–C(1)–C(5)–H(5) dihedral angle of +39.5, +39.1 and +39.6°, respectively. The conformations of the five- and six-membered ring in the three structures are essentially identical and the only difference resides in the rotation of the isopropyl moiety. Alcohol **4** had a similar conformational profile than peroxide **3**. A comparative drawing of the molecular dynamics samplings corresponding to the global and local minima of esquelane derivatives **1–4** is depicted in Fig. 3, while Table 1 shows the molecular mechanics energy values for each conformer, as well as the corresponding H(1)–C(1)–C(5)–H(5) and C(6)–C(7)–C(11)–H(11) dihedral angles. The conformations of the five- and six-membered rings in terms of the polar set of parameters proposed by Cremer and Pople,[13] which were calculated with the RICON program,[14] are also described in Table 1.

The magnitude of the $J_{1,5}$ coupling constant in esquelane derivatives **1–4** experimentally reflects the degree of mobility of the hydrindane system and the conformational population associated with such dynamic behavior. An important variation of this coupling constant was observed along the series ($J_{1,5}$=9.0 Hz for **1**, $J_{1,5}$=6.4 Hz for **2**, $J_{1,5}$=8.5 Hz for **3**, $J_{1,5}$=7.5 Hz for **4**). The calculated coupling constant for a conformation in which the dihedral angle is 0° was estimated to be 10.5 Hz, while that for a dihedral angle of +40° or −40° was estimated as 6.2 Hz. Experimentally, compounds **1**, **3** and **4** show an important contribution to the conformations wherein the H(1)–C(1)–C(5)–H(5) dihedral angle is closer to 0°, while compound **2**

Table 1. Selected dihedral angles, molecular mechanics energy and conformational parameters of esquelane derivatives **1**–**4**

Conformer	H_1–C_1–C_5–H_5[a]	C_6–C_7–C_{11}–H_{11}[a]	E_{MMX}[b]	Six-membered ring				Five-membered ring		
				Q[c]	ϕ[a]	θ[a]	Conformation	Q[c]	ϕ[a]	Conformation
1a	−41	+155	18.222	0.441	27.54	50.46	Half-chair	0.411	13.71	Between twist/envelope
1b	−40	−142	18.441	0.440	28.97	48.98	Half-chair	0.409	15.66	Distorted twist
1c	−40	+14	18.165	0.443	29.65	49.81	Half-chair	0.410	14.66	Distorted twist
1d	−28	+134	20.029	0.445	0.14	88.96	Boat	0.419	3.23	Distorted envelope
1e	−27	−147	20.240	0.426	0.52	89.83	Boat	0.420	3.80	Between envelope/twist
1f	−27	−7	20.025	0.437	0.93	88.94	Boat	0.418	4.13	Between envelope/twist
1g	−23	+150	18.906	0.402	4.68	55.86	Distorted envelope	0.411	10.12	Between twist/envelope
1h	−21	+11	18.748	0.402	7.56	56.14	Between envelope/screw-boat	0.411	11.94	Between twist/envelope
1i	−20	−134	18.956	0.414	10.56	56.34	Between envelope/screw-boat	0.410	13.31	Between twist/envelope
1j	+23	−10	19.775	0.410	1.57	54.06	Envelope	0.423	6.16	Between envelope/twist
1k	+24	+135	19.912	0.411	2.23	53.24	Distorted envelope	0.427	5.18	Between envelope/twist
1l	+31	−156	19.800	0.397	20.69	51.84	Between half-chair/envelope	0.428	4.78	Between envelope/twist
1m	+35	−134	20.236	0.572	1.39	88.06	Distorted boat	0.427	14.64	Distorted twist
1n	+35	+142	20.261	0.579	2.72	88.00	Between boat/screw-boat	0.425	14.49	Distorted twist
1o	+36	+6	20.169	0.564	1.20	88.73	Distorted boat	0.426	15.08	Distorted twist
2a	+39	+49	21.803	0.457	21.61	62.36	Between screw-boat/envelope	0.401	4.52	Between envelope/twist
2b	+40	−171	21.441	0.453	19.46	62.41	Between screw-boat/envelope	0.403	3.60	Between envelope/twist
2c	+40	−33	21.943	0.454	18.75	63.11	Between half-chair/boat	0.404	3.49	Distorted envelope
3a[d]	−44	−3	20.839	0.406	0.53	38.46	Envelope	0.424	16.72	Distorted twist
3b	−44	+29	20.864	0.407	2.20	55.84	Envelope	0.426	16.96	Distorted twist
3c	−43	+170	20.564	0.412	3.75	54.46	Distorted envelope	0.423	15.87	Distorted twist
3d	−35	+33	21.475	0.335	15.06	59.41	Between envelope/screw-boat	0.414	0.08	Envelope
3e	−31	−9	20.828	0.306	20.79	59.04	Between half-chair/boat	0.415	3.45	Distorted envelope
3f	−31	+168	20.953	0.313	21.20	60.26	Between half-chair/boat	0.414	5.28	Between envelope/twist
3g	+12	−33	24.410	0.511	4.95	80.16	Between boat/half-chair	0.418	15.98	Distorted twist
3h	+12	+27	23.968	0.491	3.80	78.81	Between boat/half-chair	0.418	16.39	Distorted twist
3i	+17	−169	24.887	0.367	17.04	75.07	Between screw-boat/boat	0.422	11.59	Between twist/envelope
3j	+41	−22	23.083	0.402	6.05	59.66	Between envelope/twist	0.424	0.06	Envelope
3k	+41	+5	21.423	0.386	2.46	57.91	Distorted envelope	0.424	0.80	Envelope
3l	+42	−161	22.572	0.382	13.28	58.69	Between envelope/screw-boat	0.428	2.92	Distorted envelope
4a[e]	−44	−3	23.024	0.403	0.23	53.49	Envelope	0.429	16.75	Distorted twist
4b	−43	+21	23.081	0.406	2.46	56.02	Distorted envelope	0.431	17.21	Twist
4c	−42	+170	23.077	0.420	6.47	55.99	Between envelope/screw-boat	0.425	14.87	Distorted twist
4d	−33	+34	23.595	0.338	17.29	61.35	Between screw-boat/envelope	0.415	1.68	Distorted envelope
4e	−31	−13	23.356	0.342	26.94	61.07	Distorted half-chair	0.410	4.01	Between envelope/twist
4f	−31	+168	23.589	0.355	24.97	62.06	Between screw-boat/envelope	0.410	5.17	Between envelope/twist
4g	+8	−173	25.800	0.471	3.32	82.27	Between boat/half-chair	0.416	12.16	Between twist/envelope
4h	+12	+45	24.765	0.486	4.80	79.87	Between boat/half-chair	0.415	16.40	Distorted twist
4i	+13	−29	25.186	0.481	7.82	80.53	Between boat/half-chair	0.416	17.96	Twist
4j	+41	−167	24.428	0.395	3.68	59.84	Distorted envelope	0.422	0.33	Envelope
4k	+41	−19	23.834	0.404	6.53	60.83	Between envelope/twist	0.420	1.30	Distorted envelope
4l	+39	+50	23.712	0.416	16.19	60.60	Between envelope/screw-boat	0.414	4.72	Between envelope/twist

[a] In deg.

[b] Molecular mechanics energy in kcal/mol.

[c] Total puckering amplitude in Å.

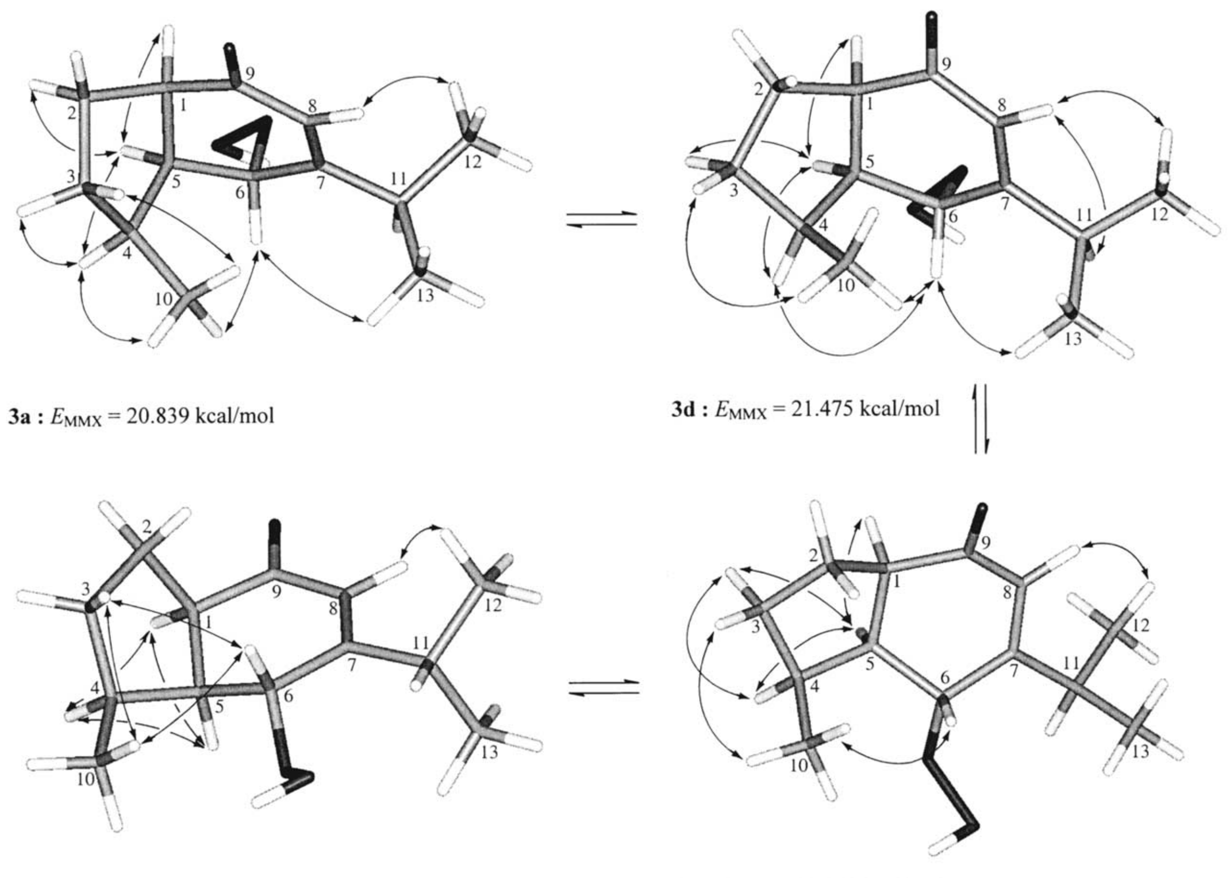
[d] Only the lowest energy conformers arising from rotation of the C_6–O–OH bonds are listed.

[e] Only the lowest energy conformers arising from rotation of the C_6–OH bond are listed.

has a more restricted flexibility and exists in conformations where the H(1)–C(1)–C(5)–H(5) dihedral angle is ca. 40°. This is in agreement with the molecular mechanics/dynamics calculations.

In order to ascertain the absolute configuration of the natural products **1** and **2**, phenol **5** was prepared by dehydration of alcohol **4** with *p*-toluenesulfonic acid in tetrahydrofuran and its optical properties compared with those of cacalol (**7**),[15] which is a sesquiterpenoid isolated from *Cacalia decomposita*,[16] of known absolute configuration. The ^{1}H NMR spectrum of **5** showed signals for two aromatic protons at δ 6.52 and 6.66 and one hydroxyl proton at δ 4.61, while the ^{13}C NMR spectrum displayed the six signals for the phenol moiety. When dehydration of **4** was carried out in EtOAc instead of tetrahydrofuran, the reaction product was ethyl ether **6**. The ^{1}H and ^{13}C NMR assignments of **5** and **6** were confirmed by DEPT and 2D NMR spectroscopy, including ^{1}H–^{1}H COSY and ^{13}C–^{1}H HETCOR.

The optical properties of a compound belonging to a closely related family of substances allow the determination of its absolute configuration, by application of the principles of antipodality and vicinity, if the absolute configuration of a member of such family is known.[17] However, this method may be applied only to related substances which further possess a very similar spatial distribution of atoms. A comparison between the minimum energy molecular mechanics structures of **5** and **7** indicated that cacalol (**7**)[15] is an adequate model to be stereochemically compared to phenol **5** due to the close structural relationship around



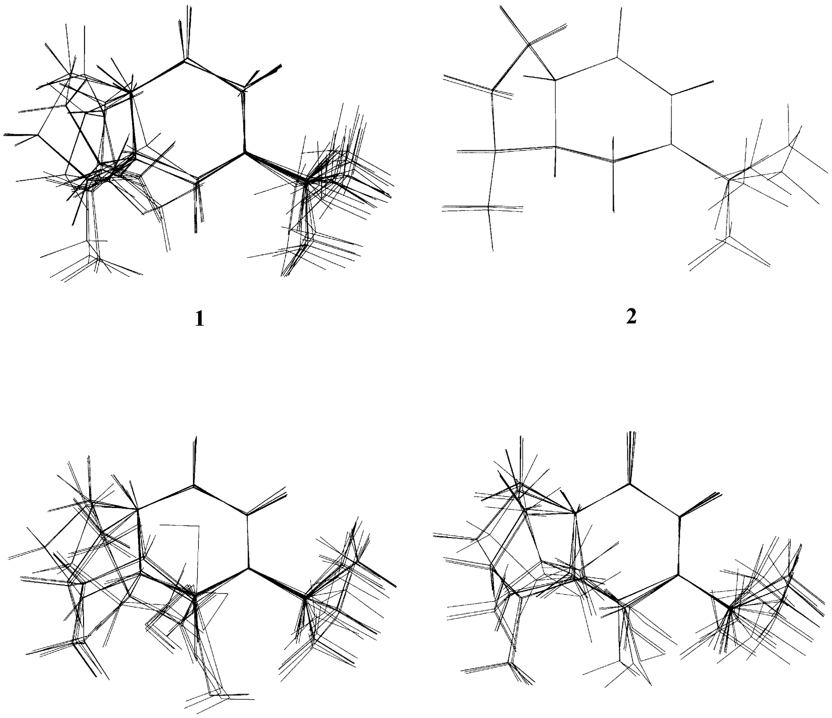
3a : $E_{\mathrm{MMX}} = 20.839$ kcal/mol

3d : $E_{\mathrm{MMX}} = 21.475$ kcal/mol

3j : $E_{\mathrm{MMX}} = 23.083$ kcal/mol

3h : $E_{\mathrm{MMX}} = 23.968$ kcal/mol

Figure 2. Representative conformers of 6-hydroperoxyesquel-7-en-9-one (**3**) and their relevant NOESY correlations.



1 **2**

3 **4**

Figure 3. Molecular dynamics samplings corresponding to the minimum energy structures of esquelane derivatives **1**–**4**.

the disymmetrically perturbed chromophore (i.e. the phenol moiety) and the analogous spatial distribution of atoms (RMS=0.17 Å, for the phenol moiety and the methyl group at the chiral center of **5** and **7**). Since both substances show a positive $[\alpha]_D$ value and CD measurements of **5** evidenced a positive Cotton effect at 271 nm, both compounds possess the same absolute stereochemistry at the sole chiral center and, consequently, the absolute configuration of compounds **1–6** can be established as depicted in Fig. 1.

The new esquelane derivatives may biogenetically derive form 6,9-guaiadiene,[18] which after oxidative cleavage of the C(9)–C(10) double bond would generate a monocyclic intermediate having an acetyl residue and an aldehyde group. Recyclization and acetyl loss would then complete the pattern. Accordingly, the bicyclic hydrocarbon skeleton for the new compounds is numbered as in guaianes.

3. Experimental

3.1. General

Column chromatography was carried out on Merck silica gel 60 (70–230 mesh ASTM) or (230–400 mesh ASTM). Optical rotations were measured in $CHCl_3$ on a Perkin–Elmer 241 polarimeter. The CD measurement was determined on a Jasco J-720 spectropolarimeter. UV spectra were measured in EtOH in a Perkin–Elmer Lambda 12 spectrometer. IR spectra were obtained in $CHCl_3$ on a Perkin–Elmer 16F PC FT spectrophotometer. NMR spectra were measured from $CDCl_3$ solutions containing TMS as the internal standard. 300 MHz ^{1}H, 75.4 MHz ^{13}C NMR and carbon–carbon connectivity spectra were registered on a Varian Associates XL-300GS spectrometer. HSQC and HMBC measurements were determined on a Varian Mercury 300 spectrometer. The 500 MHz spectra were registered on a Varian Unity Plus 500 spectrometer. LRMS were recorded on Hewlett–Packard 5989 A or Varian Saturn 2000 mass spectrometers. HRMS were measured on a VG 7070 high resolution mass spectrometer at UCR Mass Spectrometry Facility, University of California, Riverside.

3.2. Plant material

Specimens of *A. boronioides* Hook. *f.* were collected at La Hoya mountain near Esquel City, Chubut Province, Argentina, during July 1998. A voucher specimen PA N°002 is deposited at the herbarium of Facultad de Ciencias Naturales, Universidad Nacional de la Patagonia sede Esquel, where Professor Pedro E. Guerra identified the plant material.

3.3. Extraction and isolation

The essential oil (3 g, 0.6%) was obtained by hydrodistillation of the air-dried aerial parts of *A. boronioides* (500 g). The essential oil was subjected to flash chromatography,[19] using hexane–EtOAc (6:1). A total of 35 fractions were collected and monitored by TLC. Fractions 12–14 contained pure **1** (400 mg). Compound **2** was obtained as follows: fractions 18–20 were flash-chromatographed using CH_2Cl_2–EtOAc–*i*-PrOH (99:1:0.1); fractions 22–31 of this chromatography were combined and rechromatographed on an $AgNO_3$–silica gel column,[20] using hexane–EtOAc (9:1). Fractions 21–23 were flash-chromatographed using hexane–EtOAc (4:1) to afford pure **2** (10 mg).

3.3.1. (1*R*,4*S*,5*R*)-Esquel-6-en-9-one (1). Colorless oil; $[\alpha]_{589}=+18$, $[\alpha]_{578}=+18$, $[\alpha]_{546}=+21$, $[\alpha]_{436}=+39$, $[\alpha]_{365}=+73$ (*c* 2.0, $CHCl_3$); IR ($CHCl_3$) ν_{max} 2962, 2930, 1702, 1464, 1380 cm^{-1}; UV λ_{max} 221 (ε 1300), 300 (ε 160); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.49 (1H, m, H-6), 2.96 (1H, br m, H-5), 2.85 (2H, m, H-8α and H-8β), 2.76 (1H, br ddd, $J_{1,2\alpha}=J_{1,5}=9.0$ Hz, $J_{1,2\beta}=6.0$ Hz, H-1), 2.27 (1H, br septet, $J_{11,12}=J_{11,13}=6.8$ Hz, H-11), 2.12 (1H, br septet, $J_{3\alpha,4}=J_{3\beta,4}=J_{4,5}\cong6.6$ Hz, $J_{4,10}=7.1$ Hz, H-4), 2.06 (1H, m, H-2α), 1.81 (1H, m, H-2β), 1.72 (1H, m, H-3α) 1.38 (1H, m, H-3β), 1.04 (3H, d, $J_{11,12}=6.8$ Hz, Me-12), 1.03 (3H, d, $J_{11,13}=6.8$ Hz, Me-13), 0.80 (3H, d, $J_{4,10}=7.1$ Hz, Me-10); ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 212.2 (C-9), 141.3 (C-7), 118.3 (C-6), 49.9 (C-1), 47.6 (C-5), 39.1 (C-8), 38.1 (C-4), 34.6 (C-11), 32.0 (C-3), 24.9 (C-2), 21.0 (C-12), 20.8 (C-13), 15.3 (C-10); EIMS (20 eV) *m/z* (rel. int.) $[M]^+$ 192 (13), 149 (38), 136 (13), 121 (36), 107 (46), 95 (50), 93 (54), 81 (38), 79 (36), 55 (53), 43 (100); HRDEIMS *m/z* 192.1510 (calcd for $C_{13}H_{20}O$, 192.1514).

3.3.2. (1*R*,4*S*,5*S*)-Esquel-7-en-9-one (2). Colorless oil; $[\alpha]_{589}=+18$, $[\alpha]_{578}=+19$, $[\alpha]_{546}=+23$, $[\alpha]_{436}=+68$, $[\alpha]_{365}=+456$ (*c* 2.0, $CHCl_3$); IR ($CHCl_3$) ν_{max} 2960, 2928, 1650, 1466, 1376 cm^{-1}; UV λ_{max} 235 (ε 2600); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 5.85 (1H, br d, $J_{6\beta,8}=1.8$ Hz, H-8), 2.62 (1H, td, $J_{1,2\alpha}=J_{1,2\beta}=9.8$ Hz, $J_{1,5}=6.4$ Hz, H-1), 2.44 (1H, m, H-5), 2.42 (1H, br septet, $J_{11,12}=J_{11,13}=6.4$ Hz, H-11), 2.18 (1H, m, H-4), 2.13 (1H, m, H-6α), 2.06 (1H, m, H-6β), 2.02 (1H, m, H-2α), 1.90 (1H, dddd, $J_{2\alpha,3\alpha}=10.1$ Hz, $J_{2\beta,3\alpha}=4.9$ Hz, $J_{3\alpha,3\beta}=12.7$ Hz, $J_{3\alpha,4}=8.3$ Hz, H-3α), 1.74 (1H, dddd, $J_{1,2\beta}=9.8$ Hz, $J_{2\alpha,2\beta}=12.7$ Hz, $J_{2\beta,3\alpha}=4.9$ Hz, $J_{2\beta,3\beta}=11.0$ Hz, H-2β), 1.40 (1H, dddd, $J_{2\alpha,3\beta}=5.4$ Hz, $J_{2\beta,3\beta}=11.3$ Hz, $J_{3\alpha,3\beta}=12.7$ Hz, $J_{3\beta,4}=9.8$ Hz, H-3β), 1.12 (3H, d, $J_{11,12}=6.9$ Hz, Me-12), 1.11 (3H, d, $J_{11,13}=6.9$ Hz, Me-13), 0.99 (3H, d, $J_{4,10}=6.9$ Hz Me-10); ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 202.7 (C-9), 169.8 (C-7), 122.3 (C-8), 49.7 (C-1), 43.5 (C-5), 38.2 (C-4), 35.8 (C-11), 30.9 (C-3), 26.9 (C-2), 24.7 (C-6), 20.7 (C-12), 20.4 (C-13), 15.2 (C-10); EIMS (20 eV) *m/z* (rel. int.) 192 $[M]^+$ (37), 149 (38), 110 (100), 107 (18), 95 (54), 83 (12), 67 (15), 57 (9), 43 (17); HRDEIMS *m/z* 192.1507 (calcd for $C_{13}H_{20}O$, 192.1514).

3.3.3. Isomerization of compound 1 into 2. A solution of **1** (22 mg) in MeOH (2 mL) was treated with a solution of $NaHCO_3$ (11 mg) in H_2O (0.2 mL). The reaction mixture was refluxed for 30 min under an Ar atmosphere, evaporated to one-half, poured over ice, and extracted with CH_2Cl_2. The organic layer was washed with H_2O, dried with anhydrous Na_2SO_4, filtered and evaporated to dryness to give compound **2** (19 mg, 86%) identical to that described above.

3.3.4. (1*R*,4*S*,5*S*,6*R*)-6-Hydroperoxyesquel-7-en-9-one (3). A solution of **1** (220 mg) in pentane (3 mL) was stirred under an O_2 atmosphere at room temperature for 12 days. The solvent was evaporated to dryness and the residue was chromatographed eluting with hexane and increasing amounts of EtOAc (5–20%) to yield hydroperoxide **3** (57 mg, 26%) and starting material **1** (60 mg, 27%). Hydroperoxide **3** was obtained as a colorless oil; $[\alpha]_{589}=+2$, $[\alpha]_{578}=+2$, $[\alpha]_{546}=+2$, $[\alpha]_{436}=+11$, $[\alpha]_{365}=+13$ (*c* 3.0, $CHCl_3$); IR ($CHCl_3$) ν_{max} 2965, 2930, 2874, 1650, 1466, 1376 cm^{-1}; UV λ_{max} 232 nm (ε 2240); ^1H NMR ($CDCl_3$, 500 MHz) δ 8.70 (1H, br s, OOH), 6.07 (1H, br s, H-8), 4.59 (1H, d, $J_{5,6}=3.0$ Hz, H-6), 2.97 (1H, ddd, $J_{1,5}=8.5$ Hz, $J_{4,5}=7.5$ Hz, $J_{5,6}=3.0$ Hz, H-5), 2.85 (1H, ddd, $J_{1,2\alpha}=8.5$ Hz, $J_{1,2\beta}=7.0$ Hz, $J_{1,5}=8.5$ Hz, H-1), 2.73 (1H, dseptet, $J_{8,11}=1.0$ Hz, $J_{11,12}=J_{11,13}=6.5$ Hz, H-11), 2.35 (1H, dsextet, $J_{3\alpha,4}=7.5$ Hz, $J_{3\beta,4}=5.2$ Hz, $J_{4,5}=J_{4,10}=7.5$ Hz, H-4), 1.98 (1H, m, H-2α), 1.98 (1H, m, H-2β), 1.81 (1H, dddd, $J_{2\alpha,3\alpha}=9.0$ Hz, $J_{2\beta,3\alpha}=7.5$ Hz, $J_{3\alpha,3\beta}=12.5$ Hz, $J_{3\alpha,4}=7.5$ Hz, H-3α), 1.30 (1H, dddd, $J_{2\alpha,3\beta}=6.0$ Hz, $J_{2\beta,3\beta}=7.5$ Hz, $J_{3\beta,3\alpha}=12.5$ Hz, $J_{3\beta,4}=5.2$ Hz, H-3β), 1.18 (3H, d, $J_{11,12}=6.5$ Hz, Me-12), 1.16 (3H, d, $J_{11,13}=6.5$ Hz, Me-13), 0.77 (3H, d, $J_{4,10}=7.5$ Hz, Me-10), ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 202.5 (C-9), 163.9 (C-7), 127.8 (C-8), 79.4 (C-6), 46.1 (C-1), 44.8 (C-5), 36.8 (C-4), 32.2 (C-11), 32.2 (C-3), 27.0 (C-2), 22.2 (C-13), 20.8 (C-12), 16.8 (C-10); EIMS (20 eV) *m/z* (rel. int.) 224 $[M]^+$ (0.2), 206 (12), 204 (22), 190 (45), 175 (100), 162 (30), 147 (43), 133 (55), 95 (24), 81 (31), 67 (18), 43 (23); HRDCIMS (NH_3) *m/z* 225.1481 $[M+1]^+$ (calcd for $C_{13}H_{20}O_3+H$, 225.1491).

3.3.5. (1*R*,4*S*,5*S*,6*R*)-6-Hydroxyesquel-7-en-9-one (4). A solution of hydroperoxide **3** (90 mg) in MeOH (3 mL) was treated with KI (20 mg). The reaction mixture was concentrated to one-half and extracted with CH_2Cl_2. The organic layer was washed with H_2O, dried with anhydrous Na_2SO_4, filtered and evaporated under vacuum to give an oily residue (70 mg, 84%) which was purified by column chromatography. The fractions eluted with hexane–EtOAc 1:1 yielded pure **4** (20 mg, 22%) as a colorless oil; $[\alpha]_{589}=-2$, $[\alpha]_{578}=-1$, $[\alpha]_{546}=+1$, $[\alpha]_{436}=+17$, $[\alpha]_{365}=+223$ (*c* 2.0, $CHCl_3$); IR ($CHCl_3$) ν_{max} 2960, 2932, 2872, 1652, 1466 cm^{-1}; UV λ_{max} 230 (ε 2330); ^1H NMR ($CDCl_3$, 300 MHz) δ 5.89 (1H, br s, H-8), 4.36 (1H, br d, $J_{5,6}=6.0$ Hz, H-6), 2.83 (1H, br septet, $J_{11,12}=J_{11,13}=6.9$ Hz, H-11), 2.79 (1H, td, $J_{1,2\alpha}=J_{1,2\beta}=9.3$ Hz, $J_{1,5}=7.5$ Hz, H-1), 2.46 (1H, td, $J_{1,5}=J_{4,5}=7.5$ Hz, $J_{5,6}=6.0$ Hz, H-5), 2.29 (1H, br septet, $J_{3\alpha,4}=J_{3\beta,4}=J_{4,5}=J_{4,10}\approx 7.5$ Hz, H-4), 2.01 (1H, m, H-2α), 1.89 (1H, m, H-3α), 1.84 (1H, m, H2β), 1.34 (1H, m, H-3β), 1.17 (3H, d, $J_{11,12}=6.6$ Hz, Me-12), 1.16 (3H, d, $J_{11,13}=6.9$ Hz, Me-13), 1.00 (3H, d, $J_{4,11}=7.2$ Hz, Me-10), ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 201.5 (C-9), 169.7 (C-7), 123.4 (C-8), 66.0 (C-6), 51.6 (C-5), 47.4 (C-1), 37.6 (C-4), 31.8 (C-3), 31.0 (C-11), 27.5 (C-2), 22.1 (C-13), 20.9 (C-12), 17.1 (C-10); EIMS (20 eV) *m/z* (rel. int.) 208 $[M]^+$ (3), 193 (4), 180 (18), 166 (33), 165 (50), 147 (33), 137 (100), 126 (80), 119 (24), 111 (90), 83 (42), 81 (48), 55 (43), 43 (24); HRDEIMS *m/z* 208.1456 (calcd for $C_{13}H_{20}O_2$, 208.1463).

3.3.6. Phenol 5. A solution of **4** (70 mg) in THF (2 mL) was treated with *p*-toluenesulfonic acid (20 mg). The reaction mixture was refluxed during 2 min, evaporated to a small volume and extracted with CH_2Cl_2. The organic layer was washed with H_2O, dried with anhydrous Na_2SO_4, filtered and evaporated under vacuum. The residue was column chromatographed eluting with hexane–EtOAc 9:1 to yield phenol **5** (30 mg, 44%) as a pale yellow oil; $[\alpha]_{589}=+1$, $[\alpha]_{578}=+1$, $[\alpha]_{546}=+2$, $[\alpha]_{436}=+9$ (*c* 5.0, $CHCl_3$); CD $[\theta]_{270}=+1007$, $[\theta]_{226}=+2001$ (*c* 0.028 g L^{-1}, MeOH); IR ($CHCl_3$) ν_{max} 3598, 3344, 2960, 1590, 1458, 1264 cm^{-1}; UV λ_{max} 220 (ε 1000), 268 (ε 250); ^1H NMR ($CDCl_3$, 300 MHz) δ 6.67 (1H, br s, H-6), 6.52 (1H, br s, H-8), 4.49 (1H, br s, OH), 3.16 (1H, ddq, $J_{3\alpha,4}=8.0$ Hz, $J_{3\beta,4}=8.5$ Hz, $J_{4,10}=7.2$ Hz, H-4), 2.85 (1H, septet, $J_{11,12}=J_{11,13}=6.8$ Hz, H-11) 2.83 (1H, ddd, $J_{2\alpha,2\beta}=15.0$ Hz, $J_{2\beta,3\beta}=8.5$ Hz, $J_{2\beta,3\alpha}=3.7$ Hz, H-2β), 2.68 (1H, ddd, $J_{2\alpha,2\beta}=15.0$ Hz, $J_{2\alpha,3\alpha}=7.8$ Hz, $J_{2\alpha,3\beta}=8.5$ Hz, H-2α), 2.33 (1H, ddd, $J_{2\alpha,3\alpha}=7.8$ Hz, $J_{2\beta,3\alpha}=3.7$ Hz, $J_{3\alpha,3\beta}=12.3$ Hz, $J_{3\alpha,4}=8.0$ Hz, H-3α), 1.63 (1H, td, $J_{2\alpha,3\beta}=J_{2\beta,3\beta}=J_{3\beta,4}=8.5$ Hz, $J_{3\alpha,3\beta}=12.3$ Hz, H-3β), 1.28 (3H, d, $J_{4,10}=7.2$ Hz, Me-10), 1.23 (3H, d, $J_{11,12}=6.8$ Hz, Me-12), 1.23 (3H, d, $J_{11,13}=6.8$ Hz, Me-13), ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 151.5 (C-9), 151.1 (C-5), 149.6 (C-7), 126.1 (C-1), 113.8 (C-6), 110.8 (C-8), 39.8 (C-4), 34.6 (C-3), 34.1 (C-11), 26.9 (C-2), 24.1 (C-12), 24.1 (C-13), 19.9 (C-10); EIMS (20 eV) *m/z* (rel. int.) 190 $[M]^+$ (65), 175 (100), 147 (38), 133 (50), 119 (4), 105 (5), 91 (5), 43 (5); HRDEIMS *m/z* 190.1361 (calcd for $C_{13}H_{18}O$, 190.1358).

3.3.7. Ethyl ether 6. Obtained exactly as **5**, but using EtOAc instead of THF. The reaction afforded **6** (15.9 mg, 20%) as a colorless oil; $[\alpha]_{589}=+2$, $[\alpha]_{578}=+2$, $[\alpha]_{546}=+3$, $[\alpha]_{436}=+11$, $[\alpha]_{365}=+33$ (*c* 1.2, $CHCl_3$); IR ($CHCl_3$) ν_{max} 2958, 2928, 2868, 1608, 1586, 1476, 1458 cm^{-1}; UV λ_{max} 223 (ε 1590), 270 (ε 182); ^1H NMR ($CDCl_3$, 300 MHz) δ 6.68 (1H, br s, H-6), 6.54 (1H, br s, H-8), 4.05 (2H, m, OCH_2CH_3), 3.15 (1H, ddq, $J_{3\alpha,4}=8.0$ Hz, $J_{3\beta,4}=8.5$ Hz, $J_{4,10}=6.9$ Hz, H-4), 2.88 (1H, septet, $J_{11,12}=J_{11,13}=6.8$ Hz, H-11), 2.88 (1H, ddd, $J_{2\alpha,2\beta}=15.0$ Hz, $J_{2\beta,3\beta}=8.5$ Hz, $J_{2\beta,3\alpha}=3.7$ Hz, H-2β), 2.69 (1H, ddd, $J_{2\alpha,2\beta}=15.0$ Hz, $J_{2\alpha,3\alpha}=7.8$ Hz, $J_{2\alpha,3\beta}=8.5$ Hz, H-2α), 2.29 (1H, ddd, $J_{2\alpha,3\alpha}=7.8$ Hz, $J_{2\beta,3\alpha}=3.7$ Hz, $J_{3\alpha,3\beta}=12.3$ Hz, $J_{3\alpha,4}=8.0$ Hz, H-3α), 1.59 (1H, td, $J_{2\alpha,3\beta}=J_{2\beta,3\beta}=J_{3\beta,4}=8.5$ Hz, $J_{3\alpha,3\beta}=12.3$ Hz, H-3β), 1.40 (3H, t, $J=6.5$ Hz, OCH_2CH_3), 1.27 (3H, d, $J_{4,10}=6.9$ Hz, Me-10), 1.25 (3H, d, $J_{11,12}=6.8$ Hz, Me-12), 1.25 (3H, d, $J_{11,13}=6.8$ Hz, Me-13), ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 155.0 (C-9), 150.5 (C-5), 149.0 (C-7), 129.0 (C-1), 113.3 (C-6), 107.6 (C-8), 63.4 (OCH_2CH_3), 39.9 (C-4), 34.6 (C-3), 34.5 (C-11), 27.7 (C-2), 24.4 (C-12), 24.3 (C-13), 20.6 (C-10), 15.1 (OCH_2CH_3); EIMS (20 eV) *m/z* (rel. int.) 218 $[M]^+$ (83), 203 (100), 175 (52), 162 (12), 147 (17), 133 (28), 105 (5), 91 (4), 43 (5); HRDEIMS *m/z* 218.1667 (calcd for $C_{15}H_{22}O$, 218.1671).

3.4. Molecular modeling calculations

The minimum energy structures were generated using the MMX force-field as implemented in the PCModel molecular modeling program V 6.00 (Serena Software, Bloomington, IN). A systematic conformational search for the five and six-membered rings was conducted by performing dihedral angle rotations of ca. 10° in those bonds which allowed such movement, according to the corresponding Dreiding models. The π-system calculations were set for the Restricted Hartree Fock and Full Self Consistent Field options, while an implicit solvent description was applied using a dielectric constant term of 4.8 ($CHCl_3$). After the minimization procedure, the global minimum structures served as the starting point for the molecular dynamics analysis. All molecular dynamics runs were performed during 1000 ps, starting with an initial temperature of 5 K and then warmed to 298 K over a period of 300 ps. A time step for numerical integration of 1 fs was employed during all the molecular dynamics process using a viscosity term of 0.0001 cp. The heat transfer time was set at 1 fs and the equilibration time at 300 fs. Samples were saved every 1 ps resulting 1000 structures for each trajectory. The complete structure set was submitted to the Batch routine to re-minimize all the saved structures which were analyzed and classified by torsion angles employing a total energy diagram vs. time.

Acknowledgements

The authors thank CYTED (Project IV.6), Spain, and CoNaCyT, Mexico, for partial financial support. We thank María Isabel Chávez and María del Rocío Patiño Maya, Instituto de Química, UNAM, for recording the ^{1}H 500 MHz spectra and CD spectrum, respectively.

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