

A new sesquiterpenoid eremophilanolactone from *Senecio nemorensis*

Ruijian Zhao, Weidong Xie, Yuxue Liu, Fanjun Meng*, Hong Zhao and Pengxiang Lai

Marine College, Shandong University at Weihai, Weihai 264209, P. R. China

A new sesquiterpenoid eremophilanolactone and three known compounds were isolated from the MeOH extract of the aerial parts of *Senecio nemorensis* L. Their structures were elucidated on the basis of spectroscopic evidence including IR, HR-ESI-MS, 1D and 2D NMR techniques. The new compound was established as 8 α -hydroxy-6 β -(isobutanoyloxy)-eremophila-7(11),1(10)-dieno-12,8 β -lactone.

Keywords: *Senecio nemorensis* L., Compositae, sesquiterpenoid, eremophilanolactone

There are about 1200 species of the genus *Senecio* (Compositae) distributed throughout the temperate and tropical zones of the world, with more than 160 species distributed throughout China.¹ The whole plant of *Senecio nemorensis* L. has been used for the treatment of dysentery, enteritis, hepatitis, conjunctivitis, bloodshot eyes and otitis media in traditional and folk medicine.² Previous phytochemical research on *S. nemorensis* has led to the isolation of four new eremophilane sesquiterpenes and 29 known compounds.^{1–4} The results indicate that pyrrolizidine alkaloids and eremophilane sesquiterpenes are characteristic secondary metabolites.

With the aim of finding antibacterial terpenes and discovering the relationship between phytochemical constituents and ecological position, we undertook a study of the chemical constituents in roots and aerial parts of *S. nemorensis* collected in east China, and isolated five new eremophilane-type sesquiterpene lactones.^{5–7} Further studies of the aerial parts of this plant afforded a new eremophilane sesquiterpene and three known compounds. We now report their isolation and structural elucidation.

Results and discussion

The methanol extract from aerial parts of *S. nemorensis* was subjected to repeated column chromatography on silica gel to obtain a new sesquiterpene eremophilane, 8 α -hydroxy-6 β -(isobutyryloxy)-eremophila-7(11),1(10)-dieno-12,8 β -lactone (**4**), together with three known compounds: 1 β ,6 α -dihydroxy-4 α (15)-epoxyeudesmane (**1**), 1 β ,6 α -dihydroxy-4(15)-eudesmene (**2**) and 1 β ,5 α -dihydroxy-4(15)-eudesmene (**3**).

Compound **4** was obtained as colourless gum, and its molecular formula was established as C₁₉H₂₆O₅ by HR-ESI-MS (m/z = 357.1669 [M+Na]⁺, Calcd for C₁₉H₂₆O₅Na: 357.1677), revealing seven degrees of unsaturation. Its IR spectrum exhibited absorption due to hydroxy groups (3390 cm⁻¹), carbonyl groups (1766, 1742 cm⁻¹) and double bonds (1634 cm⁻¹). The ¹H and ¹³C NMR spectra of compound **4** showed the typical

signals for an isobutyryloxy group: δ_H = 2.69 (m), 1.26 (3H, d, J = 7.0) and 1.24 (3H, d, J = 7.0), and δ_C = 176.53, 34.27, 18.99 and 18.69. Apart from the above group, the ¹³C NMR and DEPT spectra revealed 15 carbon signals including three methyls, three methylenes, four methines and six quaternary carbons (including one ester carbonyl, three olefinic, and one acetal), indicating a sesquiterpene skeleton with an α,β -unsaturated γ -lactone unit. The ¹H–¹H COSY spectrum showed partial structure =CH(1)–CH₂(2)–CH₂(3)–CH(4)–CH₃(14). The HMBC spectrum showed correlations of H-1 with C-2, C-3, C-5 and C-9, of H-9 with C-1, C-5, C-7, C-8 and C-10, of H-O with C-7, C-8 and C-9, of H-6 with C-4, C-5, C-7, C-11, C-15 and C-1'. These indicated that the double bond was located between C-1 and C-10, and that the hydroxy and the isobutyryloxy group were located at C-8 and C-6, respectively. From these data and HSQC, NOESY data, the structure of compound **4** was established as 8 α -hydroxy-6 β -(isobutanoyloxy)-eremophila-7(11),1(10)-dieno-12,8 β -lactone, a new eremophilane sesquiterpene lactone.

Experimental

The IR spectra were recorded with a Bruker Vertex 70 FT-IR spectrometer as a KBr disc or film. Optical rotation was measured on a Perkin-Elmer 341 polarimeter. ¹H, ¹³C NMR (DEPT) and 2D NMR spectra were recorded on a Bruker Avance 500 spectrometer with TMS as internal reference. HR-ESI-MS spectra were obtained on Q-TOF 6510 GC/MS spectrometers using a direct insertion probe method. Silica gel (200–300 and 300–400 mesh) used for column chromatography (CC) and preparative thin layer chromatography (PTLC) precoated plates were purchased from Qingdao Marine Chemical Factory in China. Spots were detected on TLC under UV light at 254 nm and by heating after spraying with 5% H₂SO₄ in C₂H₅OH.

Plant material

The aerial parts of *Senecio nemorensis* were collected from Kunyu Mountains, Weihai, P.R. China in September 2006, and identified by associate professor Hong Zhao, (Shandong University at Weihai, Weihai). A voucher specimen (No. KY2006001) is deposited in the

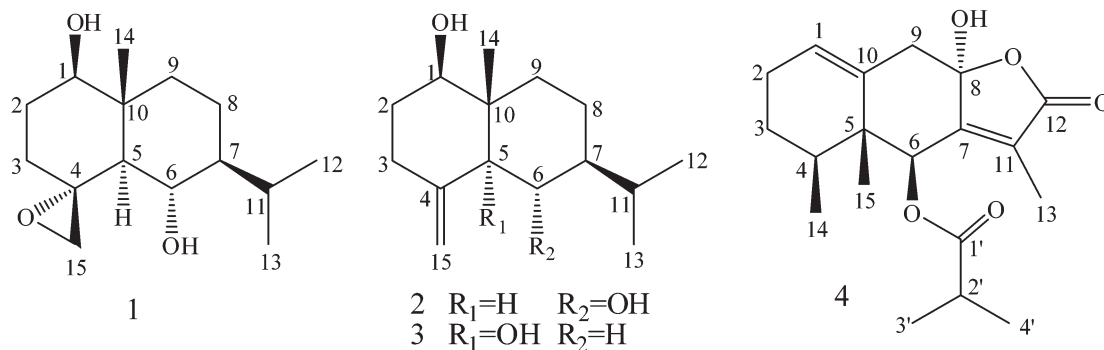


Fig. 1 Chemical structures of compounds 1–4.

* Correspondent. E-mail: mengfj@sdu.edu.cn

Table 1 ^1H , ^{13}C NMR (DEPT) and DEPT data of compounds **1–3** (CDCl_3 , δ in ppm, TMS)

	1^a		2^b		3^a	
	$\delta(\text{H})$	$\delta(\text{C})$	$\delta(\text{H})$	$\delta(\text{C})$	$\delta(\text{H})$	$\delta(\text{C})$
H-C(1)	3.41(dd, $J = 11.8, 4.4$)	78.2(d)	3.42(dd, $J = 11.3, 3.3$)	79.0(d)	4.05(dd, $J = 11.6, 5.0$)	73.1(d)
Ha-C(2)	1.53(m)	29.4(t)	1.51(m)	35.1(t)	1.83(m)	30.6(t)
Hb-C(2)	1.92(m)		1.85(dtd, $J = 12.5, 5.1, 2.2$)		1.56(m)	
Ha-C(3)	1.32(ddd, $J = 13.2, 4.7, 2.5$)	33.3(t)	2.07(td, $J = 13.4, 5.2$)	31.9(t)	2.70(m)	29.8(t)
Hb-C(3)	2.08(m)		2.33(ddd, $J = 13.2, 5.1, 2.2$)		2.15(ddd, $J = 13.7, 5.4, 1.8$)	
H-C(4)	—	61.6(s)	—	146.2(s)	—	150.6(s)
H-C(5)	1.66(d, $J = 9.7$)	49.9(d)	1.74(d, $J = 9.9$)	55.8(d)	—	76.2(s)
Ha-C(6)	3.44(t, $J = 9.7$)	67.7(d)	3.70(t, $J = 9.8$)	67.0(d)	1.58(m)	34.3(t)
Hb-C(6)	—		—		1.54(m)	
H-C(7)	1.26(m)	49.8(d)	1.29(tt, $J = 12.6, 3.0$)	49.3(d)	1.62(m)	38.3(d)
Ha-C(8)	1.22(m)	18.1(t)	1.50(m)	18.1(t)	1.51(m)	23.7(t)
Hb-C(8)	1.51(m)		1.20(qd, $J = 12.9, 2.6$)		1.24(m)	
Ha-C(9)	1.17(m)	36.9(t)	1.91(m)	36.2(t)	1.70(dd, $J = 13.0, 3.9$)	30.0(t)
Hb-C(9)	1.87(m)		1.17(m)		1.64(m)	
C(10)	—	42.0(s)	—	41.6(s)	—	42.3(s)
H-C(11)	2.24(m)	25.1(d)	2.24(m)	26.0(d)	1.48(m)	32.8(d)
Me(12)	0.92(d, $J = 6.9$)	16.0(q)	0.87(d, $J = 7.1$)	16.1(q)	0.91(d, $J = 6.7$)	20.0(q)
Me(13)	0.82(d, $J = 6.9$)	21.0(q)	0.95(d, $J = 7.1$)	21.1(q)	0.90(d, $J = 6.7$)	19.7(q)
Me(14)	0.87(s)	12.2(q)	0.70(s)	11.5(q)	0.76(s)	12.7(q)
Ha-C(15)	2.78(d, $J = 3.6$)	51.6(t)	4.75(s)	107.8(t)	4.85(s)	108.6(t)
Hb-C(15)	3.22(dd, $J = 3.6, 2.0$)		5.02(s)		4.75(s)	

^a Measured at 400 MHz for ^1H NMR and 100 MHz for ^{13}C NMR and DEPT.^b Measured at 500 MHz for ^1H NMR and 125 MHz for ^{13}C NMR and DEPT.

Laboratory of Botany, Marine College, Shandong University at Weihai.

Extraction and isolation

The powdered air-dried aerial parts of *S. nemorensis* (5.8 kg) were extracted with methanol three times at room temperature, each for 6 days. After evaporation of the combined extracts under reduced pressure, the residue (386 g) was suspended in water and then extracted successively with hexane and CHCl_3 , respectively. The CHCl_3 soluble fraction (147 g) was chromatographed on a silica-gel column (200–300 mesh, 1200 g) with an elution gradient of hexane–acetone (20:1, 10:1, 5:1). Five fractions were collected according to their TLC analysis: Fa1 to Fa5. The main constituents of Fa1 (hexane–acetone 20:1, 18.0 g) were a volatile oil and β -sitosterol (170.0 mg). Fa2 (hexane–acetone 10:1, 2.5 g) was isolated by repeated silica gel column

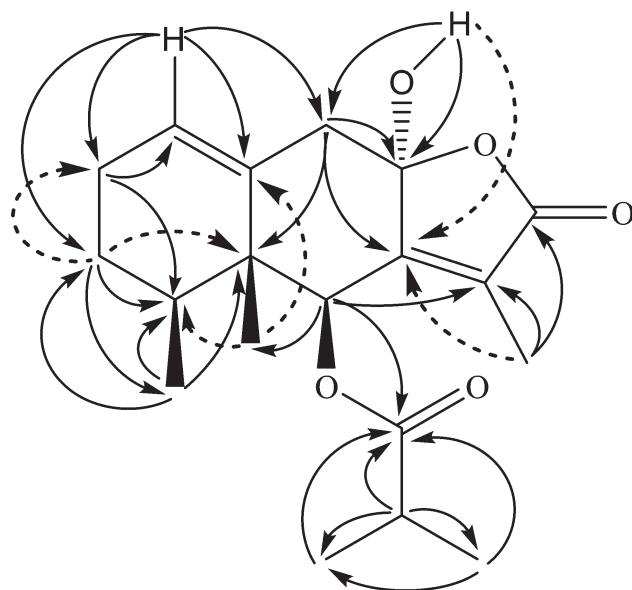
chromatography, using CHCl_3 –acetone (200:1), hexane– CHCl_3 (2:1) as eluents respectively, to yield compound **1** (5.0 mg) and a mixture (65.0 mg). The mixture was further purified by reverse phase C-18 silica column chromatography, using MeOH– H_2O (1.5:1) as eluent, to yield compound **2** (32.5 mg). Fa3 (hexane–acetone 10:1, 2.0 g) was isolated by silica gel column chromatography, using CHCl_3 –acetone (100:1), hexane–acetone (15:1–8:1–4:1) as eluents sequentially, and preparative thin layer chromatography, to yield compound **3** (19.3 mg). Fa4 (hexane–acetone 10:1, 5.0 g) was isolated by repeated silica gel column chromatography, using CHCl_3 –acetone (50:1), petroleum ether (60–90 °C)–ethyl acetate (20:1–15:1–10:1–5:1–2:1), hexane– CHCl_3 (5:1–2:1) as eluents to yield compound **4** (11.0 mg). No interesting spot was found in Fa5 (hexane–acetone 5:1, 19.0 g).

1\beta,6\alpha-dihydroxy-4a(15)-epoxyeudesmane (**1**): Colourless gum. The spot on TLC had no fluorescence under UV light at 254 nm, and

Table 2 ^1H , ^{13}C NMR and DEPT data of compound **4** (CDCl_3 , δ in ppm, TMS)

	$\delta(\text{H})$	$\delta(\text{C})$	DEPT
H-C(1)	5.80(br s)	130.3	CH
2H-C(2)	2.06(m)	24.1	CH_2
Ha-C(3)	1.57(m)	28.0	CH_2
Hb-C(3)	1.39(m)		
H-C(4)	1.78(m)	36.3	CH
C(5)	—	45.9	C
H-C(6)	5.53(d, $J = 1.6$)	78.5	CH
C(7)	—	155.6	C
C(8)	—	102.5	C
Ha-C(9)	2.75(d, $J = 13.8$)	44.7	CH_2
Hb-C(9)	2.48(d, $J = 13.8$)		
C(10)	—	133.4	C
C(11)	—	123.7	C
C(12)	—	171.2	C
Me(13)	1.85(d, $J = 1.6$)	8.4	CH_3
Me(14)	1.04(d, $J = 6.9$)	17.7	CH_3
Me(15)	0.98(s)	13.9	CH_3
C(1')	—	176.5	C
H-C(2')	2.69(m)	34.3	CH
Me(3')	1.26(d, $J = 7.0$)	19.0	CH_3
Me(4')	1.24(d, $J = 7.0$)	18.7	CH_3

Measured at 400 MHz for ^1H NMR and 100 MHz for ^{13}C NMR and DEPT.

**Fig. 2** The key HMBC correlations of compound **4**.

becomes red by heating after spaying with 5% H₂SO₄ in C₂H₅OH. $[\alpha]_D^{20} = -24.3^\circ$ (c 0.20, CH₂Cl). IR $\nu_{\max}/\text{cm}^{-1}$: 3442, 2950, 2869, 1061, 1018. The ¹H, ¹³C NMR and DEPT data were identified as *1β,6α-dihydroxy-4α(15)-epoxyeudesmane*,⁸ see Table 1.

1β,6α-dihydroxy-4(15)-eudesmene (2): Colourless needle crystal. The spot on TLC has no fluorescence under UV light at 254 nm, and becomes purple by heating after spaying with 5% H₂SO₄ in C₂H₅OH. m.p. 124–125 °C. $[\alpha]_D^{20} = +47.5^\circ$ (c 0.20, Me₂CO). IR (KBr) $\nu_{\max}/\text{cm}^{-1}$: 3400, 2934, 2870, 1646, 1464, 1457, 1386, 1060, 1003. ¹H, ¹³C NMR and DEPT data were identified as *1β,6α-dihydroxy-4(15)-eudesmene*,^{9,10} see Table 1.

1β,5α-dihydroxy-4(15)-eudesmene (3): Colourless gum. The spot on TLC has no fluorescence under UV light at 254 nm, and becomes red by heating after spaying with 5% H₂SO₄ in C₂H₅OH. $[\alpha]_D^{17} = +31.0^\circ$ (c 0.10, CH₂Cl). IR $\nu_{\max}/\text{cm}^{-1}$: 3405, 2930, 2859, 1657, 890. ¹H, ¹³C NMR and DEPT data were identified as *1β,5α-dihydroxy-4(15)-eudesmene*,^{8,11} see Table 1.

8α-hydroxy-6β-(isobutanoyloxy)-eremophila-7(11),1(10)-dieno-12,8β-lactone (4): Colourless gum. The spot on TLC has no fluorescence under UV light at 254 nm, and becomes blue by heating after spaying with 5% H₂SO₄ in C₂H₅OH. IR $\nu_{\max}/\text{cm}^{-1}$: 3390, 2976, 2932, 2880, 1766, 1742, 1634, 1469, 1460, 1385, 1188, 1150, 1126, 1012, 977, 947, 924, 909. HR-ESI-MS: *m/z*: 357.1669 ([M+Na]⁺, C₁₉H₂₆O₅Na, Calcd 357.1677). For ¹H, ¹³C NMR and DEPT data, see Table 2.

This work was supported by the Natural Science Foundation of Shandong Province of China (Y2008B50) and Excellent Scholar Youth Group Foundation (1070508200001) from Shandong University at Weihai.

Received 12 May 2010; accepted 7 June 2010

Paper 1000157 doi: 10.3184/030823410X12791286866995

Published online: 30 August 2010

References

- 1 D.Q. Fei, Z.X. Zhang, J.J. Chen and K. Gao, *Planta Med.*, 2007, **73**, 1292.
- 2 B.J. Shi, G.X. Chou and Z.T. Wang, *J. China Pharm. Univ.*, 2010, **41**, 26.
- 3 D.L. Cheng, J.J. Gao and L. Yang, *Chem. J. Chinese Univ.*, 1992, **13**, 781.
- 4 H. Wiedenfeld, S. Narantuya, D. Altanchimeg and E. Roeder, *Sci. Pharm.*, 2000, **68**, 207.
- 5 F.J. Meng, H. Zhao, W.D. Xie, R.J. Zhao, P.X. Lai, Y.X. Zhou and Y.L. Miao, *Helv. Chim. Acta*, 2007, **90**, 2196.
- 6 F.J. Meng, W.D. Xie, Kyungho Row, *J. Chem. Res.*, 2008, **7**, 376.
- 7 F.J. Meng, Y.N. Xiao, R.J. Zhao, H. Zhao, Q.L. Ma and W.D. Xie, *Nat. Prod. Res.*, 2009, **23**, 208.
- 8 K. Junichi, S. Nobuyuki, S. Mituru and W. Mitsuo, *Phytochemistry*, 2002, **59**, 811.
- 9 H.J. Zhang, G.T. Tan, B.D. Santarsiero, A.D. Mesecar, N.V. Hung, N.M. Guong, D.D. Soejarto, J.M. Pezzuto and H.H.S. Song, *J. Nat. Prod.*, 2003, **66**, 609.
- 10 J.F. Hu, S.P. Bai and Z.J. Jia, *Phytochemistry*, 1996, **43**, 815.
- 11 Z.H. Sun, B. Chen, S. Zhang and C.Q. Hu, *J. Nat. Prod.*, 2004, **67**, 1975.