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SEVEN GUAIANOLIDES FROM *EUPATORIUM CHINENSE*

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Key Word Index—*Eupatorium chinense*; Compositae; new sesquiterpene lactone; new guaianolides; hydroperoxides; ^{1}H and ^{13}C NMR; photo-oxygenation.

Abstract—Two allyl hydroperoxy guaianolide sesquiterpene lactones (peroxyeupahakonin-A and -B) and five new guaianolides (eupahakonin-A and -B, eupahakonenin-A and -B, and eupahakonesin) were isolated from *E. chinense* and characterized. The allyl hydroperoxy sesquiterpene lactones were characterized by spectral and chemical methods. They were prepared chemically by photosensitized oxygenation of eupahakonin-A.

INTRODUCTION

Previous studies on sesquiterpene lactones from the genus *Eupatorium* have yielded several germacranolides, heliangolides and guaianolides. For example, the four heliangolides eupalinin-A, -B, -C, and -D were isolated from *E. lindleyanum* [1]. Among these compounds, eupalinin-A showed significant inhibitory against KB cell culture. Eupachifolin-A, which is a *cis,cis*-4,5,9,10-germacradienolide possessing an α,β-unsaturated aldehyde group, and the related guaianolides the eupachifolins-B, -C, -D and -E were obtained from *E. chinense* var. *simplicifolium* [2]. Peroxysachalinin, sachalinin, and sachalin were obtained from *E. sachalinense*, along with a known compound, eupatoriopicrin, which is the main component of this plant[3]. In the present paper, we describe the isolation and structure determination of two new guaianolide hydroperoxides and five closely related guaianolides from *E. chinense* L. var. *hakonense* (Nakai) Kitam. The structure and stereochemistry of the new sesquiterpene lactones were determined by chemical transformations and extensive applications of ^{1}H and ^{13}C NMR spectrometry.

RESULTS AND DISCUSSION

Peroxyeupahakonin-A (**1**, $C_{20}H_{24}O_8$ mp 143–146° $[\alpha]_D$ −165°) and -B (**2**, $C_{20}H_{24}O_8$ mp 147–148° $[\alpha]_D$ +35.4°), and eupahakonin-A (**3**, $C_{20}H_{24}O_7$ oil $[\alpha]_D$ −146°) and -B (**4**, $C_{20}H_{24}O_7$ oil $[\alpha]_D$ +65.3°) were isolated by repeated column chromatography on Si gel and then separated by prep. TLC. The IR, UV, and ^{1}H NMR spectra of each of these guaianolides supported the presence of an α,β-unsaturated γ-lactone system and an ester group. Characteristic of this class of compounds a *trans* diaxial relationship between the protons at C-5–C-6 and C-6–C-7 was observed in the signal for H-6 which occurred as a well-defined one-proton doublet of doublets at δ 4.50 (J = 8.5, 11 Hz), δ 4.52 (J = 9, 10 Hz), δ4.46 (J = 8.5, 11 Hz), and δ 4.46 (J = 9, 10 Hz) in **1**, **2**, **3**, and **4**, respectively (Table 1).

In the ^{13}C NMR spectra (Table 2) of **1** and **2**, seven oxygenated carbon atoms were observed; two carbonyls, two singly protonated, two doubly protonated, and one unprotonated. The peaks at δ 94.6 in **1** and δ 95.4 in **2** were uniquely characteristic of an allylic carbon (C-1) bearing a hydroperoxy group. Furthermore, both compounds gave a positive ferrous thiocyanate test [4]. Acetalization of **1** and **2** with Me_2CO and *p*-TsOH gave the acetonides **1a**: oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3550, 3300, 1775 and 1715; *m/z* 432 $[M]^+$ and **2a**: oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3550, 3350, 1770, and 1715; *m/z* 432 $[M]^+$, respectively. These results indicated that **1** and **2** contained the same ester group. In the ^{1}H NMR spectrum of **2**, the exocyclic methylene protons gave two singlets at δ 5.03 and 5.09, instead of the broad singlet due to C-10 vinyl methyl protons seen in **1**. Treatment of **1** with triphenylphosphine afforded an alcohol, which was identical in all respects with eupahakonin-A (**3**). Peroxyeupahakonin-B (**2**) was likewise converted to eupahakonin-B (**4**).

Alkaline hydrolysis of **3** with 10% KOH in aqueous dioxane, followed by acetylation with acetic anhydride and pyridine gave the acetate **3a**: IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600, 1765, 1740 and 1665; *m/z* 304 $[M]^+$, which was also formed by the same reactions from **4**.

From the above physical data and experiments, it was clear that peroxyeupahakonin-A (**1**) and -B (**2**), as well as eupahakonin-A (**3**) and -B (**4**), were regioisomers of the double bond at C-10, and that eupahakonin-A (**3**) and -B (**4**) are reduction products of peroxyeupahakonin-A (**1**) and -B (**2**), respectively. The configuration of the hydroperoxy group at C-1 and the ester group at C-8 in **1** was determined as follows. The chemical shift of H-5 in **3** (δ 2.85) clearly indicated that the hydroxyl group was α, because of the δ 0.51 high field shift compared with that of H-5 in **1** (δ 3.36). On the other hand, α-orientation of H-8 was supported by the fact that 8.5% NOE was observed between H-7 and H-8. From the above physical data and experiments, the structures of peroxyeupahakonin-A, -B, eupahakonin-A, and -B were established as **1**, **2**, **3**, and **4**, respectively.

Eupahakonenin-A (**5**, $C_{20}H_{24}O_6$, oil, $[\alpha]_D$ −95°) and -B (**6**, $C_{20}H_{24}O_6$, oil, $[\alpha]_D$ +22°) were purified several times by prep. TLC. These new sesquiterpene lac-

Table 1. ^{1}H NMR data of compounds 1–7 (100 MHz, TMS as internal standard)

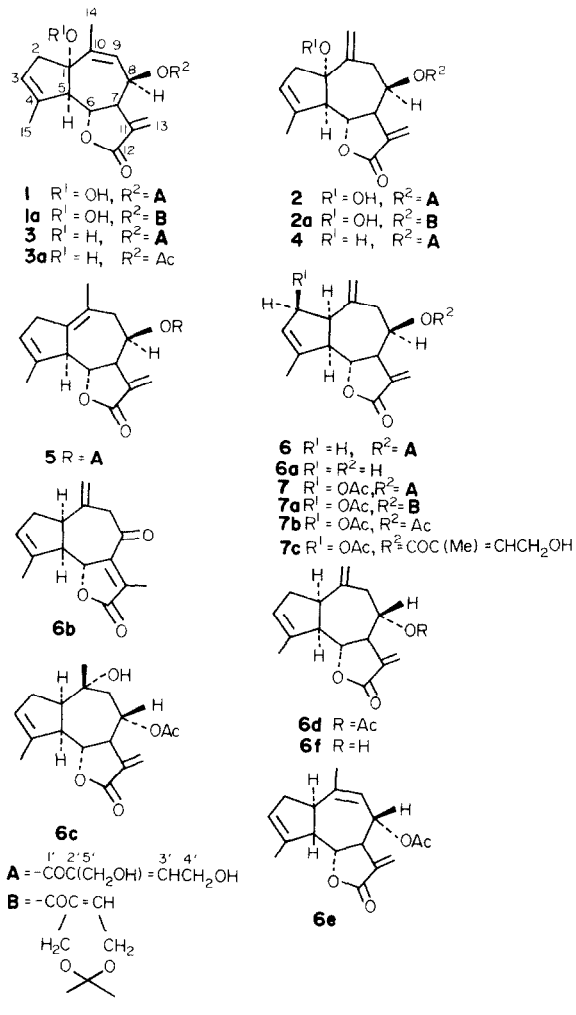
Compound	H-1	H-2	H-3	H-5	H-6	H-7	H-8	H-9	H-13	H-14	H-15	Miscellaneous
1†	—	2.58 *m*	5.40 *s*(*br*)	3.36 *d* (11)	4.50 *dd* (8.5;11)	3.59 *m*	5.88 *dd*(*br*) (3;6.3)	5.70 *dd*(*br*) (1;6.3)	5.57 *d*(3) 6.11 *d*(3.4)	.92 *s*(*br*)	1.92 *s*(*br*)	6.75 *t*(5.8,H-3'), 4.20 *s*(H-5') 4.30 *d*(5.8,H-4')
1a†	—	2.36 *m*	5.40*	3.36 *d*(*br*) (11)	4.48 *dd* (8.5; 11)	3.52 *m*	5.88 *dd*(*br*) (3; 6.3)	5.70 *d*(*br*) (6.3)	5.57 *d*(3) 6.15 *d*(3.4)	1.92 *s*(*br*)	1.92 *s*(*br*)	6.78 *m*(H-3'), 4.24–4.42 *m* (H-4',5'), 1.36 *s*(2 × Me)
2‡	—	2.90*	5.60*	2.90*	4.52 *dd* (9; 10)	3.40 *m*	5.60*	2.49 *dd*(3.2; 14.5) 3.14 *dd*(3.9; 14.5)	5.65 *d*(3.2) 6.18 *d*(3.7)	5.03 *s* 5.09 *s*	1.84 *s*(*br*)	6.80 *t*(5.8, H-3'), 4.27 *s*(H-5'), 4.35 *d*(5.8, H-4')
2a†	—	2.58*	5.58 *s**	2.85*	4.24*	3.60 *m*	5.58*	2.47 *dd*(3.2; 14) 3.12 *dd*(3.9; 14)	5.58 *d*(3) 6.10 *d*(3.7)	5.02 *s* 5.08 *s*	1.80 *s*(*br*)	6.79 *m*(H-3'), 4.24–4.54 *m* (H-4', 5'), 1.35 *s*(2 × Me)
3†	—	2.56 *m*	5.58 *s*(*br*)	2.85 *s*(*br*) (11)	4.46 *dd* (8.5; 11)	3.60 *m*	5.88 *dd*(*br*) (3; 6)	5.48 *m*)	5.56 *d*(3.2) 6.10 *d*(3.8)	1.92 *m*	1.96 *s*(*br*)	6.77 *t*(5.8, H-3'), 4.22 s(H-5'), 4.35 *d*(5.8,H-4')
3a†	—	2.44 *m* 2.90 *m*	5.58*	2.77 *d*(*br*) (10.5)	4.35 *dd* (8.5; 10.5)	3.36 *m*	5.48*	2.32 *dd*(5; 14) 3.05 *dd*(5; 14)	5.51 *d*(3) 6.26 *d*(3.8)	4.94 *s* 5.16 *s*	1.89 *s*(*br*)	2.01 *s*(OAc)
4†	—	2.62 *m* 2.75 *m*	5.60*	2.86 *d*(*br*) (10)	4.46 *dd* (9; 10)	3.59 *m*	5.60 *d**	2.36 *dd*(3.5; 14) 3.19 *dd*(3.9; 14)	5.58 *d*(3.5) 6.10 *d*(3.8)	4.91 *s* 5.07 *s*	1.81 *s*(*br*)	6.80 *t*(5.8, H-3'), 4.22 s(H-5'), 4.32 *d*(5.8, H-4'),
5§	—	3.03 *s**	5.57*	3.40 *d*(*br*) (10)	4.12 *t* (10)	3.06 *m*	5.60 *m**	2.61 *m*	5.44 *d*(3) 6.15 *d*(3.5)	1.61 *s*	1.97 *s*(*br*)	6.80 *t*(5.8, H-3'),4.31 *s*(H-5'), 4.38 *d*(5.8, H-4')
6§	3.17*	2.48 *m*	5.53*	2.81 *t*(*br*) (9)	4.48 *dd* (9; 10.5)	3.17*	5.60 *m**	2.55 *d* (5)	5.52 *d*(3) (6.22 *d*(3.8)	4.86 *s* 4.98 *s*	1.84 *s*(*br*)	6.79 *t*(5.8, H-3'), 4.28 *s*(H-5') 4.35 *d*(5.8, H-4')
6a§	3.14*	2.42 *m*	5.52*	2.77 *t*(*br*) (9)	4.38 *dd* (9; 10.5)	2.96 *m*	4.32 *m*	2.50 *d* (5)	5.56 *d*(3) 6.33 *d*(3.8)	4.90 *s* 4.97 *s*	1.84 *s*(*br*)	—
6b§	3.27*	2.50*	5.59 *m*	2.50*	4.95 *dd* (2; 10.5)	—	—	3.37 *d*(*br*)(10) 3.50 *d*(*br*)(10)	2.18 *d* (2)	5.06 *s*	1.90 *s*(*br*)	—
6d§	3.24*	2.36 *m*	5.49 *m*	2.71*	4.03 *dd* (9; 11)	3.10 *m*	4.92 *m*	2.27 *dd*(6; 14.5) 2.70 *dd*(5; 14.5)	5.59 *d*(3) 6.18 *d*(3.8)	4.90 *s* 5.04 *s*	1.85 *s*(*br*)	2.14(OAc) —
6e§	3.01*	2.40*	5.50 *m*	2.85*	4.06 *dd* (9; 11)	3.04 *m*	5.34 *m*	5.26 *s*(*br*)	5.66 *d*(3) 6.22 *d*(3.8)	1.81 *s*	1.92 *s*(*br*)	2.15(OAc) —
6f§	3.27 *q* (8)	2.37 *m*	5.48 *m*	2.75*	3.99 *dd* (9; 11)	2.75*	3.90 *m*	2.26 *dd*(6; 14.5) 2.75 *m*	6.13 *d*(3) 6.23 *d*(3.8)	4.91 *s* 5.03 *s*	1.85 *s*(*br*)	—
7§	3.46 *dd* (6; 8)	5.64*	5.64*	2.69*	4.64 *dd* (9; 10)	3.23 *m*	5.64*	2.69*	5.54 *d*(3) 6.31 *d*(3.5)	5.06 *s* 5.11 *s*	2.02 *s*	6.85 *t*(5.8, H-3'), 4.32 *s*(H-5'), 4.39 *d*(5.8, H-4'), 2.0 *s*(OAc)
7a§	3.44 *dd* (6; 8)	5.57*	5.67 *s*(*br*)	2.60*	4.57 *dd* (9; 10)	3.20 *m*	5.57*	2.60*	5.49 *d*(3) 6.31 *d*(3.5)	5.03 *s* 5.09 *s*	2.01 *s*	6.75 *m*(H-3'), 4.35; 4.44 *m*(H-4', 5'), 2.01 *s*(OAc), 1.44 *s*(2 × Me)
7b§	3.42 *dd* (6; 8)	5.51*	5.62 *s*(*br*)	2.50*	4.59 *dd* (9; 10)	3.13 *m*	5.51*	2.50*	5.48 *d*(3) 6.28 *d*(3.5)	5.00 *s* 5.06 *s*	1.98 *s*	1.97 *s*(2 × OAc)
7c§	3.42 *dd* (6; 8)	5.58*	5.64 *s*(*br*)	2.72*	5.59 *dd* (8.5; 11)	3.18 *m*	5.56*	2.53 *dd*(8; 14) 2.79 *dd*(6.5; 14)	5.48 *d*(3) 6.26 *d*(3.8)	5.03 *s* 5.08 *s*	1.99 *s*	6.67 *t*(5.8, H-3'), 4.32 *d*(5.8, H-4'),

Figures in parentheses are coupling constants in Hz.
*Signal partially obscured or superimposed.
†d_6–Me_2CO.
‡CD_3OD.
§$CDCl_3$.

Table 2. ^{13}C NMR data of compounds **1–7** (25.05 MHz, TMS as internal standard)*

	1 (d_6-Me_2CO)	**2** C_5D_5N)	**3** (d_6-Me_2CO)	**4** (d_6-Me_2CO)	**5** ($CDCl_3$)	**6** ($CDCl_3$)	**7** ($CDCl_3$)	**6d** ($CDCl_3$)	**6e** ($CDCl_3$)
1	94.6 *s*	95.4 *s*	83.4 *s*	84.5 *s*	125.2 *s*	47.6 *d*	50.6 *d*	47.7 *d*	47.5 *d*
2	44.2 *t*	43.3 *t*	47.7 *t*	47.8 *t*	37.9 *t*	37.3 *t*	80.2 *d*	36.6 *t*	37.9 *t*
3	123.4 *d*	125.6 *d*	123.6 *d*	125.4 *d*	125.4 *d*	126.6 *d*	126.1 *d*	126.3 *d*	125.6 *d*
4	142.7 *s*†	144.5 *s*†	145.5 *s*†	147.0 *s*†	139.9 *s*†	139.6 *s*†	147.9 *s*†	140.9 *s*†	138.2 *s*†
5	60.7 *d*	59.8 *d*	66.4 *d*	64.8 *d*	56.4 *d*	56.4 *d*	55.8 *d*	56.7 *d*	55.1 *d*
6	78.9 *d*	79.3 *d*	79.0 *d*	79.7 *d*	79.9 *d*	80.0 *d*	80.0 *d*	80.2 *d*	79.5 *d*
7	48.2 *d*	48.5 *d*	48.2 *d*	47.8 *d*	55.3 *d*	48.5 *d*	47.9 *d*	49.3 *d*	46.8 *d*
8	67.9 *d*	68.0 *d*	67.8 *d*	68.7 *d*	66.7 *d*	68.2 *d*	68.3 *d*	74.2 *d*	73.1 *d*
9	122.8 *d*	37.1 *t*	119.6 *d*	36.7 *t*	37.6 *t*	40.3 *t*	39.0 *t*	38.4 *t*	121.6 *d*
10	141.7 *s*†	137.5 *s*†	142.2 *s*†	139.2 *s*†	136.7 *s*†	143.1 *s*†	138.7 *s*†	142.6 *s*†	142.7 *s*†
11	135.6 *s*	135.5 *s*	135.9 *s*	136.3 *s*	135.2 *s*	134.4 *s*†	133.7 *s*†	137.0 *s*†	136.6 *s*†
12	169.0 *s*	169.3 *s*	169.0 *s*	169.1 *s*	169.1 *s*	169.4 *s*	169.1 *s*	169.6 *s*	169.7 *s*
13	121.6 *t*	121.5 *t*	121.2 *t*	121.1 *t*	119.5 *t*	122.0 *t*	122.5 *t*	122.0 *t*	122.9 *t*
14	24.4 *q*	118.1 *t*	24.9 *q*	115.8 *t*	24.1 *q*	116.4 *t*	120.1 *t*	116.8 *t*	27.7 *q*
15	17.8 *q*	16.9 *q*	17.9 *q*	17.1 *q*	17.7 *q*	16.7 *q*	17.2 *q*	17.2 *q*	17.6 *q*
1′	166.4 *s*	166.4 *s*	166.5 *s*	166.2 *s*	165.9 *s*	165.8 *s*	165.8 *s*	—	—
2′	131.8 *s*	132.1 *s*	132.0 *s*	132.2 *s*	131.1 *s*	131.3 *s*	131.1 *s*	—	—
3′	145.9 *d*	145.8 *d*	145.9 *d*	145.9 *d*	144.1 *d*	143.9 *d*	144.0 *d*	—	—
4′	59.2 *t*	58.7 *t*	59.2 *t*	59.1 *t*	58.7 *t*	58.8 *t*	58.8 *t*	—	—
5′	56.9 *t*	56.3 *t*	57.0 *t*	56.9 *t*	56.7 *t*	56.9 *t*	56.8 *t*	—	—
Ac(C=O)	—	—	—	—	—	—	170.1 *s*	168.9 *s*	169.0 *s*
Ac(CH_3)	—	—	—	—	—	—	21.3 *q*	21.2 *q*	21.2 *q*

*Assignment established by single frequency off-resonance decoupling.
†Assignments may be interchanged.



tones were similar to eupahakonin-B (**4**) on the basis of the IR, UV, and mass spectral data, except for the absence of a *tert*-hydroxyl group. Extensive double irradiation ^{1}H NMR experiments of **5** and **6** established the steric arrangements of the portion consisting of C-6–C-7. Further, the ^{1}H NMR spectrum of **5** was similar to that of **6**, except for the presence of the vinyl–methyl group and the absence of exocyclic methylene protons, indicating that these compounds might be regioisomers of the double bond at C-10. The stereochemistry at C-1 and the ester group at C-8 in **6** was determined as follows. Hydrolysis of **6** with 10% KOH in aqueous dioxane, followed by oxidation of the resulting alcohol (**6a**, mp 144–146°, *m/z* 246 $[M]^+$) with pyridinium dichromate [5] gave the butenolide **6b** (oil, mass spectrum *m/z* 244 $[M]^+$).

Dehydration of cumambrin-A (**6c**), isolated from *Crysanthemum ornatum* var. *spontaneum* [6], with $SOCl_2$ in pyridine gave **6d** (mp 89–91°) and **6e** (mp 101–102.5°). On hydrolysis with 10% KOH in aqueous dioxane, the acetate **6d** afforded the alcohol **6f** (mp 125–130° (decomp.), *m/z* 246 $[M]^+$), the ^{1}H and ^{13}C NMR spectra of which were almost superimposable on those of **6a** except for the configuration at C-8. Oxidation of **6f** with pyridinium dichromate gave a compound, which was identical with the butenolide **6b**. Hence, the stereochemistry of H-1 and H-8 of eupahakonenin-B (**6**) was α. From the above results, the structures of eupahakonenin-A and -B were established as **5** and **6**, respectively.

Furthermore, photo-oxygenation of **5** in MeOH with visible light and methylene blue as sensitizer afforded peroxyeupahakonin-A (**1**) and -B (**2**), thereby confirming the absolute structure of (**5**).

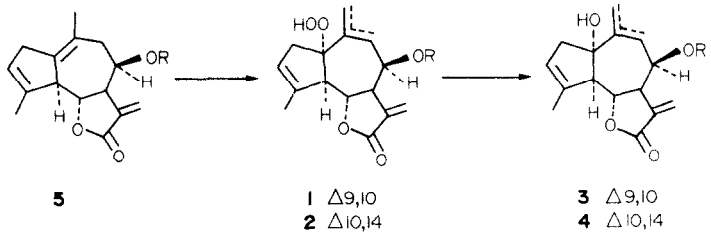
Eupahakonesin (**7**, oil, $[\alpha]_D$ −49.5°) gave rise to a molecular ion at *m/z* 418, in agreement with molecular formula $C_{22}H_{26}O_8$. Treatment of **7** with *p*-TsOH in acetone afforded the acetonide **7a** (oil, *m/z* 458 $[M]^+$). In addition, alkaline hydrolysis of **7** with 10% Na_2CO_3 in aqueous dioxane, followed by acetylation gave the diacetate **7b** (oil, *m/z* 346 $[M]^+$). From the above experiments and physical data (see Experimental and Tables 1 and 2), this new guaianolide had the same absolute stereostructure as eupahakonenin-B (**6**) except for the additional presence of an acetoxyl group [IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3450 (OH), 1770 (γ-lactone), 1735 (OAc and ester), and 1660 (C=C); ^{1}H NMR (Table 1) and ^{13}C NMR (Table 2)]. Further, the ^{1}H NMR spectrum of **7** was very similar to that of eupachifolin-C (**7c**) which was isolated from *E. chinense* var. *simplicifolium* [2], except for the ester group. Hence, the structure of eupahakonesin was represented by **7**.

The co-occurrence of peroxyeupahakonin-A (**1**) and -B (**2**), and eupahakonin-A (**3**), -B (**4**), and eupahakonenin-A (**5**) in this plant is strongly indicative of a biosynthetic interrelationship between these compounds. A sequence of biogenetically feasible steps is shown in Scheme 1.

EXPERIMENTAL

Mps are uncorr. ^{1}H and ^{13}C NMR spectra were measured at 100 and 25.05 MHz, respectively. MS, direct inlet, 70 eV. $[\alpha]_D$, UV, CD, and ORD in MeOH. TLC and prep. TLC: precoated Si gel plates (0.25 mm) F_{254}; spots detected by UV light (254 nm) and by spraying with 1% $CeSO_4$–10% H_2SO_4.

Extraction and separation. The MeOH extract of a fresh whole plant (680 g) of *E. chinense* var. *hakonense* collected



Scheme 1. Biosynthetic relationships of compounds **1**–**5**.

at Toyone, Aichi prefecture, Aug. 1979, was divided into the *n*-hexane- (10.9 g), $CHCl_3$- (4.9 g) and EtOAc-soluble fractions (3.1 g). The crude $CHCl_3$ extract was chromatographed on Si gel using a $CHCl_3$–Me_2CO gradient (6:1 starting mixture). 50-ml fractions were collected: fraction 1 (148.5 mg; $CHCl_3$–Me_2CO, 6:1), fraction 2 (336.8 mg; $CHCl_3$–Me_2CO, 6:1), fraction 3 (556 mg; $CHCl_3$–Me_2CO, 4:1) and fraction 4 (148.5 mg; $CHCl_3$–Me_2CO, 2:1). Fraction 2 was purified on a smaller Si gel column using $CHCl_3$–MeOH (14:1) as solvent. The main fractions (220 mg) showed a single spot; prep. TLC (10% $AgNO_3$–Si gel, C_6H_6–*iso*-PrOH, 6:1) gave eupahakonenin-A (**5**) (66.7 mg) and -B (**6**) (76.6 mg). Fraction 3 was rechromatographed on Si gel using a $CHCl_3$–MeOH gradient to give the less polar fractions (232.5 mg) and the more polar fractions (108.7 mg), respectively. The less polar fraction yielded eupahakonesin (**7**) (81 mg), while the more polar fraction was purified by prep. TLC (10% $AgNO_3$–Si gel; C_6H_6–*iso*-PrOH, 6:1) to afford peroxyeupahakonin-A (**1**) (41.4 mg) and -B (**2**) (48.4 mg), respectively. Fraction 4 was rechromatographed on Si gel using a $CHCl_3$–MeOH gradient to give two major products (32.3 mg) which ran together on TLC. These were purified several times by prep. TLC (10% $AgNO_3$–Si gel; C_6H_6–$CHCl_3$–Me_2CO, 3:2:2) to give pure eupahakonin-A (**3**) (7 mg) and -B (**4**) (3.3 mg), respectively.

Peroxyeupahakonin-A (**1**). $C_{20}H_{24}O_8$; mp 143–146° (amorph., Et_2O–EtOAc); $[\alpha]_D^{26}$ −165° (MeOH; *c* = 0.21); UV λ_{max}^{MeOH} nm: 209 (ε 16,491); IR ν_{max}^{KBr} cm^{-1}: 3450, 1760, 1715, 1660; CD curve: $[\theta]_{255}$ −3378, $[\theta]_{295}$ 0; ORD (*c*0.19): negative plane curve.

Peroxyeupahakonin-B (**2**). $C_{20}H_{24}O_8$; mp 147–148° (colorless needles, Me_2CO); $[\alpha]_D^{26}$ +35.4° (MeOH; *c*0.18); UV λ_{max}^{MeOH} nm: 210 (ε 16000); IR ν_{max}^{KBr} cm^{-1}: 3420–3250, 1765, 1715, 1655; CD curve: $[\theta]_{222}$ 0, $[\theta]_{228}$ +1664, $[\theta]_{237}$ 0, $[\theta]_{260}$ −1821, $[\theta]_{290}$ 0; ORD (*c*0.2): $[\phi]_{360}^{T}$ $+22\times10^2$, $[\phi]_{335}^{P}$ $+28\times10^2$, $[\phi]_{310}^{T}$ $+46.2\times10^2$, $[\phi]_{280}^{T}$ $+76.4\times10^2$.

Eupahakonin-A (**3**). $C_{20}H_{24}O_7$; colourless oil; $[\alpha]_D^{25}$ −146° (*c*0.2, MeOH); UV λ_{max}^{MeOH} nm: 210 (ε 16200), 300 (ε 243); IR ν_{max}^{KBr} cm^{-1}: 3400–3450, 1760, 1710, 1660; CD curve: $[\theta]_{247}$ −2575 $[\theta]_{257}$ −3277, $[\theta]_{292}$ 0; ORD (*c*0.2): negative plane curve.

Eupahakonin-B (**4**). $C_{20}H_{24}O_7$; colourless oil; $[\alpha]_D^{25}$ +65.3° (MeOH; *c*0.2); UV λ_{max}^{MeOH} nm: 212 (ε 14395); IR ν_{max}^{KBr} cm^{-1}: 3430, 1760, 1715, 1660; CD curve: $[\theta]_{210}$ −11,660, $[\theta]_{224}$ 0, $[\theta]_{229}$ +1167, $[\theta]_{234}$ 0, $[\theta]_{246}$ −1944, $[\theta]_{259}$ −2334, $[\theta]_{288}$ 0; ORD (*c*0.2): $[\phi]_{361}^{T}$ 10.8×10^2, $[\phi]_{335}^{P}$ $+15\times10^2$, $[\phi]_{311}^{T}$ 12.2×10^2, $[\phi]_{280}^{T}$ $+16\times10^2$.

Eupahakonenin-A (**5**). $C_{20}H_{24}O_6$; colourless oil; $[\alpha]_D^{25}$ −95° (MeOH; *c*0.2); UV λ_{max}^{MeOH} nm: 210 (ε 16146); IR $\nu_{max}^{CHCl_3}$ cm^{-1}; 3450, 1775, 1715, 1655; MS *m/z*: 360 $[M]^+$, 342, 246, 228; CD curve: $[\theta]_{222}$ −40230, $[\theta]_{250}$ −8382, $[\theta]_{267}$ 0, $[\theta]_{278}$ +587, $[\theta]_{295}$ 0; ORD (*c*0.19): negative plane curve.

Eupahakonenin-B (**6**). $C_{20}H_{24}O_6$: colourless oil; $[\alpha]_D^{25}$ +22° (MeOH; *c*0.19); UV λ_{max}^{MeOH} nm: 210 (ε 15692); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3450, 1770, 1715, 1645; MS *m/z*: 360 $[M]^+$, 342, 246, 228; CD curve: $[\theta]_{216}$ −10903, $[\theta]_{230}$ 0, $[\theta]_{260}$ −2019, $[\theta]_{285}$ 0; ORD (*c*0.2): $[\phi]_{361}^{T}$ $+8.9\times10^2$, $[\phi]_{335}^{P}$ $+12.8\times10^2$, $[\phi]_{310}^{T}$ $+18.3\times10^2$, $[\phi]_{280}^{P}$ $+13.5\times10^2$.

Eupahakonesin (**7**). $C_{22}H_{26}O_8$: colourless oil; $[\alpha]_D^{20}$ −49.5° (MeOH; *c*0.2); UV λ_{max}^{MeOH} nm: 211.5 (ε 14209); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3450, 1770, 1735, 1660; MS *m/z*: 418 $[M]^+$, 400, 376, 358, 340, 304, 262, 244, 226; CD curve: $[\theta]_{244}$ −7511.

Acetalization of **1**. To a soln of 8.6 mg **1** in 2 ml Me_2CO was added 5 mg *p*-TsOH, and the soln stirred for 4 hr at room temp. The soln was neutralized with $NaHCO_3$ and poured into ice-H_2O. The CH_2Cl_2 extract was washed with H_2O, dried over Na_2SO_4, and then evapd. The crude product was purified through a small Si gel column to give **1a** (7.3 mg): colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3550, 3300, 1775, 1715, 1660; MS *m/z* 432 $[M]^+$, 417, 401, 374, 358, 340, 264, 260, 244, 226.

Treatment of **1** *with triphenylphosphine*. A MeOH soln of **1** (23.9 mg) was treated with triphenylphosphine (24.4 mg) at room temp. for 1 hr. Removal of the solvent gave a gummy material which was purified on Si gel column to afford a pure compound (17.2 mg) which was identical in all respects with eupahakonin-A (**3**).

Acetalization of **2**. An Me_2CO soln of **2** (12.4 mg) was treated with *p*-TsOH (5 mg) with stirring for 3 hr at room temp. Work-up as **1a** gave **2a** (10.9 mg): colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3550, 3350, 1770, 1715, 1662; MS *m/z*: 432 $[M]^+$, 416, 401, 374, 358, 340, 260, 244, 226.

Treatment of **2** *with triphenylphosphine*. **2** (28.9 mg) was treated with triphenylphosphine in the same manner as **1** to afford eupahakonin-B (**4**) (16 mg).

Hydrolysis of **3**. To a soln of **3** (17.2 mg) in dioxane (2 ml) was added 10% KOH soln (4 ml). The reaction mixture was stirred for 6 hr at room temp. and after the usual work-up, the crude product was purified by prep. TLC to give a pure sesquiterpene lactone (1.7 mg) as a colourless oil. The lactone was acetylated with Ac_2O (0.5 ml) and pyridine (1 ml) for 24 hr at room temp. The usual work-up afforded the monoacetate **3a** (0.6 mg): colourless oil; UV λ_{max}^{MeOH} nm: 213 (ε 4023); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600, 1765, 1740, 1665, 1635; CD curve: $[\theta]_{217}$ −10,462, $[\theta]_{246}$ −666, $[\theta]_{260}$ −913, $[\theta]_{295}$ 0; MS *m/z*: 304 $[M]^+$, 262, 244. 3a was also obtained from **4** by the same set of reactions.

Hydrolysis of **6**. A soln of **6** (54.4 mg) in dioxane (5 ml) was treated with 10% KOH–dioxane (3:1, 5 ml) at room temp. for 3 hr. After the usual work-up, the crude product was separated by prep. TLC ($CHCl_3$–MeOH, 9:1) to give **6a**: colourless needles (28.5 mg), mp 144–146° (Et_2O); UV λ_{max}^{MeOH} nm: 208 (ε 8764); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3620, 1768, 1670, 1645; MS *m/z*: 246 $[M]^+$; 228; CD curve: $[\theta]_{210}$ 0, $[\theta]_{220}$ −20048, $[\theta]_{254}$ −802, $[\theta]_{260}$ −859, $[\theta]_{290}$ 0.

Oxidation of **6a**. **6a** (13.2 mg) was stirred with pyridinium dichromate (60.3 mg) in dry CH_2Cl_2 (5 ml) at room temp. for 7 hr. After addition of Et_2O. The reaction mixture and filtered through a Celite pad and the filtrate concd *in vacuo*. The crude product was purified by prep. TLC to give the butenolide **6d** (0.6 mg): colourless oil; UV λ_{max}^{MeOH} nm (ε): 206 (4502), 243 (5348); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1765, 1695, 1645; MS *m/z*: 244 $[M]^+$.

Dehydration of **6c**. To a pyridine soln of **6c** (100 mg) was added $SOCl_2$ (116.3 mg) at 0° and the mixture allowed to stand for 1.5 hr. Work-up as usual gave a crude oil which showed two spots on TLC. It was purified by prep. TLC (*n*-hexane–EtOAc, 9:1) to afford **6d** (16.8 mg) and **6e** (35.5 mg). **6d**: colourless needles, mp 89–91° (petrol–Et_2O); $[\alpha]_D^{23}$ +139.5° ($CHCl_3$; *c*0.2); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1770, 1740, 1665, 1645; MS *m/z*: 288 $[M]^+$, 228. **6e**: colourless needles, mp 101–102.5° (*iso*-Pr_2O–Et_2O); $[\alpha]_D^{23}$ +75° ($CHCl_3$; *c*0.2); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1775, 1740, 1665; MS *m/z*: 288 $[M]^+$, 244, 228.

Hydrolysis of **6d**. **6d** (16 mg) was treated with 10% KOH in dioxane under the same conditions as **6** to give the alcohol **6f** (6 mg): colourless needles, mp 125–130° (decomp. Et_2O); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600, 1765, 1665, 1640; MS *m/z*: 246 $[M]^+$, 228.

Oxidation of **6f**. **6f** (6 mg) was oxidized with pyridinium dichromate in dry CH_2Cl_2 under the same condition as **6a** to afford a sesquiterpene lactone (0.3 mg) whose physical and spectral data were identical to those of **6b**.

Photo-oxygenation of **5** to give **1** and **2**. A soln of **5**

(55.1 mg) and methylene blue (1.14 mg) in MeOH (13 ml) was irradiated internally with a high-pressure Hg lamp (Riko UVL-400P with Pyrex filter) in a H_2O-cooled immersion apparatus through which O_2 was circulated for 25 min. Removal of the solvent gave an oil which was purified by column chromatography on Si gel ($CHCl_3$–MeOH, 9:1). The fraction which was positive to the ferrous thiocyanate test was purified by repeated prep. TLC (10% $AgNO_3$–Si gel, C_6H_6–*iso*-PrOH, 6:1) to afford peroxyeupahakonin-A (**1**) (3.9 mg) and -B (**2**) (5.8 mg).

Acetalization of **7**. Me_2CO soln of **7** (4.9 mg) was treated with *p*-TsOH under the same condition as **1** to afford the acetonide **7a** (5.1 mg): colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1770, 1720–1735, 1660; MS *m/z*: 458 $[M]^+$, 416, 400, 398, 358, 340, 322, 244, 227.

Hydrolysis of **7**. **7** (81.8 mg) was treated with 10% Na_2CO_3–dioxane (3:1, 10 ml) soln at room temp. for 12 hr. Work-up as usual gave an oil which was purified by prep. TLC to give a pure alcohol, which was then acetylated by the usual manner to afford the diacetate **7b** (3.3 mg): colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1765, 1735, 1660; MS *m/z*: 346 $[M]^+$, 304, 286, 261, 244, 226.

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