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DITERPENES AND PHLORACETOPHENONES FROM *EUPHORBIA PORTULACOIDES*

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Key Word Index—*Euphorbia portulacoides*, Euphorbiaceae; diterpenes; lathyranes; abietanes; phloracetophenones.

Abstract—Chemical investigation of *Euporbia portulacoides* afforded, in addition to several known metabolites, i.e. two kauranes, three abietane lactones and four phloracetophenones, one new kaurane, four new lathyranes and two new abietane acids related to co-occurring lactones.

INTRODUCTION

The genus *Euphorbia* is the largest in the spurge family. This vast genus of over 1000 species is subdivided into many subgenera and sections, a number of which have been treated as distinct genera [1]. Detailed study of the profile of all secondary metabolites could contribute to taxonomic subdivision of this complex genus [2–4]. Most of the representatives are characterized by the occurence of highly irritant latex [5]. The irritant properties are due to diterpenes of the macrocyclic type mainly with a tigliane or ingenane skeleton [6]. *E. portulacoides*, a native of Chile, has not been investigated previously. In the popular medicine it has been used as a purgative with properties similar to those of *E. lathyris* [7, 8].

RESULTS AND DISCUSSION

An extract of the whole plant of *E. portulacoides* L. contained, in addition to some widespread triterpenes (Experimental), four new lathyranes (**1–4**), three known abietane lactones (**5** [9], **6** [10, 11] and **7** [9, 12]), two new methyl abietanates (**8** and **9**), the known **10** [13] and the new kaurane **11**, the known ingenane (**12**) [14] and four known phloracetophenones (**13–16**).

The ^{1}H NMR and ^{13}C NMR spectra of **1–4** showed similarities to each other differing mainly in the signals for the ester residues (Tables 1 and 2). While **1** was obviously a tetraacetate in **2–4** an acetate group was replaced by a isobutyrate, methylbutyrate or benzoate group, respectively. By spin decoupling two sequences: (i) H-1 through H-3 with a secondary methyl group at C-2, and (ii) H-5 through H-12 with methyl groups at C-6 and C-12 were determined. A cyclopropane moiety involving C-9 through C-11 was recognized from the upfield shifted signals in both the ^{1}H- and ^{13}C-NMR spectra (Tables 1 and 2). The remaining quartenary carbon signals assignable to a keto group and an epoxide must be the connecting centres for the two sequences. Their connections were determined from the long range correlations observed in a HMBC experiment. Table 3 summarizes the results obtained with the benzoate **4** (analogous results were obtained with **1–3**). The ^3J correlations between both H-12 and H-20 with the singlet at δ207.3 placed the keto group at C-14 and consequently the epoxide ring between C-4 and C-15. A series of correlations between both C-4 and C-15 and the surrounding hydrogens via two or three bonds (Table 3) confirmed the structure, which despite the stereochemistry corresponds to that of ingol polyester. However, the spectral data of **1** differ from those of ingol tetraacetate [15]. The largest differences were visible in the chemical shifts of the signals for the five-membered ring. Thus an isomer has to be assumed. The NOE results revealed that we were dealing with a 2-*epi*-ingol tetraester. In particular, the strong effect between H-16 and H-3, which itself strongly interacts with H-5, supported this proposal. All relevant NOE effects, listed in Table 3, are in excellent agreement with the energy minimized conformation calculated using PCMODEL (Scheme 1). The relative position of the ester residues followed from the long range correlations between the proton at ester-bearing carbons and the respective carbonyl group. A literature survey for ingol polyesters revealed only compounds with ingol configuration. From a *Euphorbia* species, Marco *et al.* [personal communication] obtained 2,3-bis-*epi*-ingols. Table 4 summarizes the diagnostic data for each of the isomers.

Table 1. ^{1}H NMR data of compounds **1**–**4** (CDCl$_3$, 400 MHz; int. standard, residual CHCl$_3$ signal = 7.26 ppm)

H	**1**	**2**	**3**	**4**	*m*
1(2H)	2.09	2.09	2.09	2.08	*d*
2	1.89	1.87	1.86	1.85	*ddq*
3	5.03	5.05	5.06	4.98	*d*
5	5.61	5.57	5.60	5.71	*q*
7	4.97	5.04	5.14	5.29	*d*
8	4.57	4.57	4.58	4.67	*dd*
9	1.32	1.27	1.22	1.41	*dd*
11	1.06	1.08	1.09	1.12	*dd*
12	4.85	4.86	4.86	4.90	*dd*
13	2.92	2.92	2.92	2.98	*dq*
16	1.03	1.03	1.03	0.98	*d*
17	2.07	2.08	2.08	2.16	*d*
18	1.11	1.10	1.09	1.13	*s*
19	0.84	0.85	0.85	0.88	*s*
20	1.04	1.05	1.05	1.06	*d*
3-OAc	2.03	2.01	2.00	2.02	*s*
8-OAc	2.05	2.05	2.04	2.02	*s*
12-OAc	2.10	2.10	2.10	2.11	*s*
7-OR	2.16 *s*	2.65 *qq*	2.46 *ddq*	8.05 AA′	
		1.22 *d*	1.73 *ddq*	7.49 BB′	
		1.20 *d*	1.51 *ddq*	7.60 C	
			0.93 *t*		
			1.19 *d*		

J (Hz): 1, 2 = 9; 2, 3 = 9; 2, 16 = 7; 5, 17 = 1; 7, 8 = 1.5; 8, 9 = 11; 9, 11 = 9; 11, 12 = 11; 12, 13 = 4; 13, 20 = 7.5; OiBu: 2, 3 = 2, 4 = 7; OMebu: 2, 3_1 = 2, 3_2 = 2, 5 = 3, 4 = 7; 3_1, 3_2 = 14.

Table 2. ^{13}C NMR data of compounds **1**, **4** and **9** (CDCl$_3$, 100 MHz; int. standard, solvent peak = 77.0 ppm)

C	**1**	**4**	**9**
1	31.1 *t*	31.2 *t*	39.3 *t*
2	30.9 *d*	31.3 *d*	18.3 *t*
3	80.5 *d*	80.3 *d*	41.5 *t*
4	69.9 *s*	70.1 *s*	33.1 *s*
5	117.1 *d*	117.2 *d*	54.2 *d*
6	139.4 *s*	139.2 *s*	19.2 *t*
7	77.2 *d*	77.3 *d*	41.0 *t*
8	71.0 *d*	71.5 *d*	69.5 *s*
9	25.0 *d*	25.2 *d*	61.3 *d*
10	19.4 *s*	19.4 *s*	37.8 *s*
11	30.7 *d*	30.8 *d*	71.7 *d*
12	70.7 *d*	70.7 *d*	197.5 *s*
13	42.9 *d*	43.1 *d*	136.9 *s*
14	207.3 *s*	207.3 *s*	151.4 *d*
15	71.4 *s*	71.4 *s*	137.3 *s*
16	16.1 *q*	16.1 *q*	*
17	17.3 *q*	17.4 *q*	128.8 *t*
18	29.0 *q*	29.2 *s*	33.8 *q*
19	16.1 *q*	16.1 *q*	21.9 *q*
20	13.4 *q*	13.4 *q*	17.8 *q*
OMe			52.2 *q*
OAc	170.8 *s*	170.5 *s*	
	21.0 *q*	21.0 *q*	
	170.4 *s*	170.4 *s*	
	21.0 *q*	21.0 *q*	
	170.2 *s*	170.3 *s*	
	21.0 *q*	20.6 *q*	
	170.0 *s*		
	20.6 *q*		
OBz		130.1 *s*	
		129.6 *d*	
		128.4 *d*	
		133.2 *d*	
		165.4 *s*	

*Obscured.

In addition to diterpenes of the macrocyclic type, two new abietanes (**8** and **9**) were obtained. The structures followed from the ^{1}H NMR spectral data (Table 5) and by analogy to the co-occurring lactones which, according to Scheme 2, are possible biogenetic precursors. Attempted transformation of the lactone **7** to the methyl ester **9** failed. Under acidic condition in the presence of methanol the complete deterioration of the starting material was observed and no traces of **9** were detected, thus eliminating the artificial nature of these two metabolites. The selected ^{1}H NMR data are listed in Table 5. Those for the A ring were overlapping multiplets and are thus omitted. Thus ^{13}C NMR data of **9** (Table 2) support the structure.

The ^{1}H NMR spectral data of **11** (Table 5) were similar to those of the co-occurring **10** isolated previously from *Euphorbia sieboldiana* [13]. An additional acetate signal at δ2.12 and the downfield shift of H-17 pointed to the structure. Acetylation of **10** afforded a product with spectral data identical to those of **11** confirming the structure.

The ^{1}H NMR data of the known phloroacetophenones **13**–**16** are listed in Table 6 as this data is not available in some of the earlier investigations.

The results with *E. portulacoides* showed again that the diterpenes are of chemotaxonomic relevance for the genus. Though the variation is not broad, the co-occurence of diterpenes originating from different biogenetic pathways may be of additional value. Ingoles were described from different Euphorbiaceae [6]. The chemotaxonomic relevance of phloroacetophenones is not clear and needs confirmation.

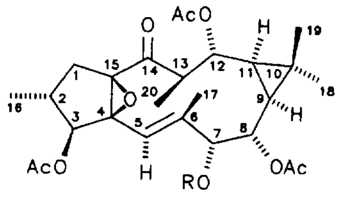
EXPERIMENTAL

Air-dried whole plant material (collected on 25 January 1994 in Lenca VIII Region, Chile; herbarium specimen deposited at the University of Concepcion, Chile) was extracted at room temp. with petrol–MTB (methyl-*tert*-butyl ether)–MEOH (1:1:1). After removal of waxy material by treatment with MeOH at −20°, the filtrate was evpd and sepd by solvent partition (petrol/10% aq. MeOH). From the crude NMR spectrum of the petrol fr. (1 g) mainly triterpenes comprising lupeol, cycloartenol and euphol and a large quantity of triglycerides were recognized. Open-column reversed-phase-chromatography

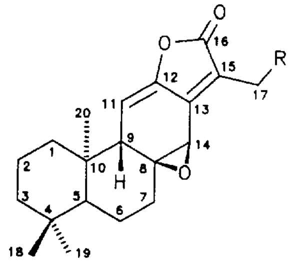
Table 3. NOE effects and long range correlations with **4**

H	NOE	H	HMBC
2	3 (2)	1	4 (m); 15 (m); 3 (w)
3	5 (3); 2 (1); 16 (s)	2	1 (m); 3 (s)
5	$2/6_{Bz}$ (3); 7 (2); 3 (7); 13 (1.5); 9 (6)	3	16 (s); 2 (m); $C{=}O_{Ac}$ (s)
7	$2/6_{Bz}$ (1); 5 (2); 8 (7); 17 (vs)	5	17 (vs); 4 (w); 15 (s); 6 (m)
8	7 (4): 12 (3); 13 (1); 19 (w)	7	17 (m); 9 (s); 8 (s); 5 (s); 6(s); $C{=}O_{Bz}$ (w)
12	8 (3); 13 (4); 19 (m)	8	9 (vs); 7 (vw); $C{=}O_{Ac}$ (s)
13	5 (1); 12 (7); 8 (2); 17 (s); 20 (s)	9	10 (w); 18 (vs); 8 (s); 7 (m); 12 (s)
16	3 (5);	11	19 (s); 10 (s); 9 (s)
17	7 (10); 8 (3.5); 13 (7)	12	11 (s); $C{=}O_{Ac}$ (s); 14 (vs)
18	9 (10); 19 (s)	13	20 (m); 14 (w)
19	12 (9); 8 (6); Ac (s); 18 (s)	16	2 (vs); 3 (vs)
20	12 (3)	17	7 (s); 5 (s); 6 (s)
$OBz_{2/6}$	5 (1); 7 (< 0.5); 9 (1)	18	19 (s); 10 (s); 11 (s)
		19	10 (s); 9 (m); 18 (s); 11 (s)
		20	13 (vs); 12 (vs); 14 (s)
		12-OAc	170.3 ($C{=}O_{Ac}$) (s)
		3-OAc	170.4 ($C{=}O_{Ac}$) (s)
		8-OAc	170.5 ($C{=}O_{Ac}$) (s)
		$OBz_{2/6}$	165.4 ($C{=}O_{Bz}$) (vs)

Abbreviations: v = very; w = weak; m = medium; s = strong.

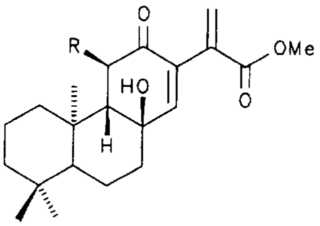


	1	2	3	4
R	Ac	iBu	Mebu	Bz

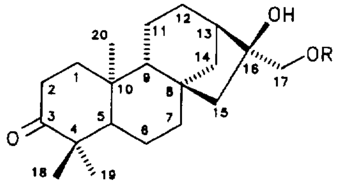


	5	6	7
R	H	OH	H

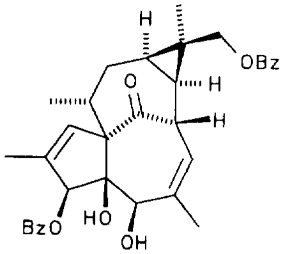
11β,12β-epoxide



	8	9
R	H	OH



	10	11
R	H	Ac



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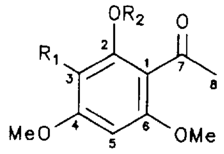
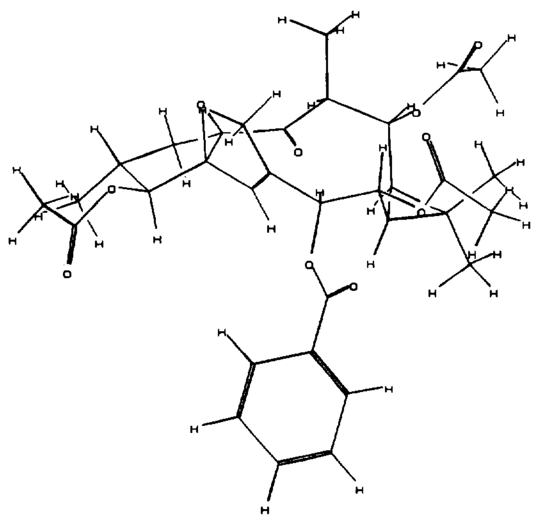


	13	14	15	16
R_1	H	H	Me	Me
R_2	H	Me	H	Me

of the MeOH fraction (RP8, 30 × 100 mm) with mixts of MeOH and H_2O gave 4 frs. Fr. 1 ($MeOH{-}H_2O$ 1:1) contained carbohydrates and other non-terpenoid polar compounds which were not further characterized. Fr. 2 ($MeOH{-}H_2O$ 3:2), 3 ($MeOH{-}H_2O$ 4:1) and 4 (MeOH) were combined and sepd by MPCC (medium pressure CC) with mixts of petrol–MTB–MeOH of increasing polarity. The resulting seven frs were combined



Scheme 1. Calculated conformation of **4**.

Table 4. Diagnostic ^{1}H NMR data of tetraacetates of ingol, 2-*epi*-ingol and 2,3-bis-*epi*-ingol ($CDCl_3$)

H	Ingol tetraacetate*	2-*epi*-Ingol tetraacetate	2,3-bis-*epi*-Ingol tetraacetate†
1	2.78 *dd* (15, 9)	2.09 *d* (9)	2.25 *dd* (13.5, 10.5)
1′	1.70 *d* (15)	2.09 *d* (9)	2.00 *dd* (13.5, 7)
2	2.47 *m*	1.89 *m*	2.10 *m*
3	5.34 *d* (8.5)	5.03 *d* (9)	5.28 *d* (5.5)
7	5.09 *d* (1)	4.97 *d* (1.5)	5.19 *br s*
16	0.97 *d* (7.5)	1.03 *d* (7)	0.92 *d* (7)

*Taken from ref. 13.

†Personal communication from Prof. Marco.

Table 5. Selected ^{1}H NMR data of compounds **8**, **9** and **11** ($CDCl_3$, 400 MHz; int. standard, residual $CHCl_3$ signal = 7.26 ppm)

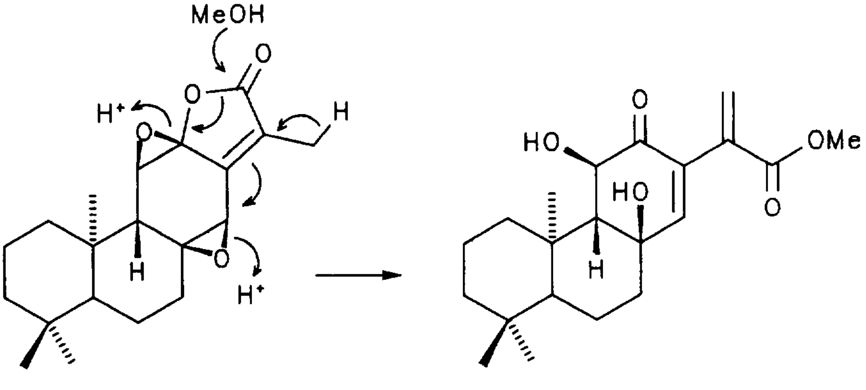
H	**8**	**9**	H	**11**
5	0.99 *br d*	1.10 *d*	2	2.48 *dd*
9	1.92 *br d*	2.01 *br s*	9	1.21 *br d*
11	2.93 *dd*	4.23 *br d*	13	2.06 *br s*
11′	2.59 *br d*		17	4.05 *d*
14	6.66 *d*	6.79 *d*	17′	3.91 *d*
17	6.26 *d*	6.34 *d*	18	1.08 *s*
17	5.72 *d*	5.89 *d*	19	1.08 *s*
18	0.92 *s*	0.93 *s*	20	1.02 *s*
19	0.79 *d*	0.81 *s*	OAc	2.12 *s*
20	0.76 *s*	0.79 *s*		
OMe	3.73 *s*	3.74 *s*		

J (Hz): Compounds **8** and **9**: 8ax, 14 = 17, 17′ = 1; compound **8**: 5, 6ax = 13; 9, 11 = 6.5; 11, 11′ = 18; compound **9**; 6, 6ax = 12; 9, 11 = 1; compound **11**: 1, 2 = 6.5; 1′, 2 = 9; 9, 11 = 9; 17, 17′ = 11.

Table 6. ^{1}H NMR data of compounds **13**–**16** ($CDCl_3$, 400 MHz; int. standard, residual $CHCl_3$ signal = 7.26 ppm)

H	**13**	**14**	**15**	**16**	m
3	6.06 *d* (2)	6.09 *s*			
5	5.92 *d* (2)	6.09 *s*	5.96 *s*	6.25 *s*	
8	2.61	2.45	2.62	2.49	*s*
Me			2.01	2.06	*s*
OH	14.04		13.97		*s*
	3.86	3.82	3.90	3.85	*s*
	3.82	3.78	3.89	3.82	*s*
		3.78		3.71	*s*

into three (A–C). After addition of MeOH to fr. A, 7 mg **7** crystallized. The filtrate sepd by HPLC ($MeOH–H_2O$ 3:2; always RP 18; 250 × 8 mm) gave 1 mg **16** (R_t 6.7 min), 3 mg **13** (R_t 9 min) and two mixts which were further sepd by TLC (petrol–EtOAc 4:1). Mixt. 1 gave 2 mg **1** (R_f 0.33) and 2 mg **15** (R_f 0.47). Mixt. 2 gave 1 mg **4** (R_f 0.39), 4 mg **5** (R_f 0.56) and a mixt. which gave after HPLC ($MeOH–H_2O$ 7:3) 2 mg **2** (R_t 18.2 min) and 2 mg **3** (R_t 22.5 min). From fr. B, a further 6 mg of **7** crystallized. The filtrate sepd by TLC (petrol–EtOAc 3:1) gave two



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Scheme 2.

bands which were further sepd by HPLC (MeOH–H_2O 7:3). Band 1 gave 1 mg **13** (R_t 5.8 min) and 1 mg **5** (R_t 10.3 min). Band 2 gave 3 mg **1** (R_t 5.7 min), 2 mg **7** (R_t 6.8 min) and 1 mg **12** (R_t 10.7 min). HPLC (MeOH–H_2O 3:2) of fr. C gave 3 mg **14** (R_t 4.8 min), 5 mg **10** (R_t 7.6 min), 6 mg **4** (R_t 21.0 min) and 4 mixts which were further sepd by TLC. Mixt. 1 (petrol–EtOAC 7:3) gave 5 mg **11** (R_f 0.34), mixt. 2 (petrol–EtOAc 3:2) 4 mg **9** (R_f 0.21), mixt. 3 (petrol-EtOAc 13:7) 2 mg **1** (R_f 0.81) and mixt. 4 (petrol–MTB 1:1) 3 mg **8** (R_f 0.2) and 3 mg **6** (R_f 0.3).

Known compounds were identified by comparing their spectral data with those of authentic material or with literature data.

2-epi-*Ingol*-3,7,8,12-*tetraacetate* (**1**). IR $\nu_{\max}^{\mathrm{CHCl_3}}$: 1740, 1250 (ester), 1720 (ketone); MS *m/z* (rel. int.): 534.247 $[M]^+$ (100) (calc. for $C_{28}H_{38}O_{10}$ 534.247), 474 $[M-AcOH]^+$ (29), 432 $[474-\text{ketene}]^+$ (27), 414 $[474-AcOH]^+$ (7) 372 $[432-AcOH]^+$ (23), 354 $[414-AcOH]^+$ (7), 312 $[372-AcOH]^+$ (22), 294 $[354-AcOH]^+$ (18).

2-epi-*Ingol*-3,8,12-*triacetate*-7-*isobutyrate* (**2**). IR $\nu_{\max}^{\mathrm{CHCl_3}}$: 1740, 1250 (ester), 1720 (ketone); MS *m/z* (rel. int.): 562.278 $[M]^+$ (51) (calc. for $C_{30}H_{42}O_{10}$ 562.278), 502 $[M-AcOH]^+$ (12), 432 $[502-(CH_3)_2C{=}C{=}O]^+$ (13), 372 $[432-AcOH]^+$ (23), 312 $[372-AcOH]^+$ (22), 71 $[(CH_3)_2CHCO]^+$ (100).

2-epi-*Ingol*-3,8,12-*triacetate*-7-*methylbutyrate* (**3**). IR $\nu_{\max}^{\mathrm{CHCl_3}}$: 1740, 1250 (ester), 1720 (ketone); MS *m/z* (rel. int.): 576.294 $[M]^+$ (65) (calc. for $C_{31}H_{44}O_{10}$ 576.294), 516 $[M-AcOH]^+$ (15), 492 $[M-CH_3CH_2(CH_3)C{=}C{=}O]^+$ (7), 474 $[M-\text{MebuOH}]^+$ (5), 432 $[492-AcOH]^+$ (28), 372 $[432-AcOH]^+$ (22), 312 $[372-AcOH]^+$ (17), 85 $[CH_3CH_2(CH_3)CHCO]^+$ (100).

2-epi-*Ingol*-3,8,12-*triacetate*-7-*benzoate* (**4**). IR $\nu_{\max}^{\mathrm{CHCl_3}}$: 1740, 1707, 1240 (ester, ketone); MS *m/z* (rel. int.): 596.262 $[M]^+$ (11) (calc. for $C_{33}H_{40}O_{10}$ 596.262), 536 $[M-AcOH]^+$ (2), 494 $[536-\text{ketene}]^+$ (1), 476 $[536-AcOH]^+$ (1), 372 $[494-PhCOOH]^+$ (1), 354 $[476-PhCOOH]^+$ (1), 312 $[372-AcOH]^+$ (2), 105 $[PhCO]^+$ (100).

Methyl-8β-*Hydroxy*-12-*oxo-ent-abietadi*-13,15(17)-*ene*-16-*oate* (**8**). MS *m/z* (rel. int.): 346.214 $[M]^+$ (20) (calc. for $C_{21}H_{30}O_4$), 328 $[M-H_2O]^+$ (11), 314 $[M-MeOH]^+$ (12), 286 (14), 190 (34), 69 (79), 55 (100).

Methyl-8β,11β-*Dihydroxy*-12-*oxo-ent-abietadi*-13, 15(17)-*ene*-16-*oate* (**9**). MS *m/z* (rel. int.): 362.209 $[M]^+$ (6) (calc. for $C_{21}H_{30}O_5$ 362.209), 344 $[M-H_2O]^+$ (9), 329 $[344-CH_3]$ (5), 312 $[344-MeOH]^+$ (8), 226 (21), 218 (6), 57 (100).

17-*Acetoxy*-16β-*hydroxy*-3-*oxo-ent-kaurane* (**11**). MS *m/z* (rel. int.): 362.246 $[M]^+$ (5) (calc. for $C_{22}H_{34}O_4$ 362.246), 344 $[M-H_2O]^+$ (6), 289 $[M-CH_2OAc]^+$ (100), 258 (8), 247 (10), 229 (12), 203 (13), 95 (44).

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