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CLERODANE AND *ENT*-HALIMANE DITERPENES FROM *POLYALTHIA LONGIFOLIA*

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Key Word Index—*Polyalthia longifolia*; Annonaceae; clerodane; *ent*-halimane; bicyclic diterpene.

Abstract—A hexane extract of the stem bark of *Polyalthia longifolia* furnished nine new clerodane and *ent*-halimane diterpenes, i.e. 16-hydroxycleroda-4(18),13-dien-16,15-olide, 16-oxocleroda-4(18),13*E*-dien-15-oic acid, cleroda-4(18),13-dien-16,15-olide, 16-hydroxy-*ent*-halima-5(10),13-dien-16,15-olide, 16-oxo-*ent*-halima-5(10),13*E*-dien-15-oic acid, *ent*-halima-1(10),13*E*-dien-16,15-olide, 16-oxo-*ent*-halima-5(10),13*E*-dien-15-oic acid, *ent*-halima-5(10),13-dien-16,15-olide and *ent*-halima-5(10),13*E*-dien-16,15-olide, along with five known clerodane diterpenes. The structures of these compounds were elucidated by spectroscopic methods. The 13*E* configuration of two of the new *ent*-halimanes and one of the known clerodanes was firmly established by NOE experiments.

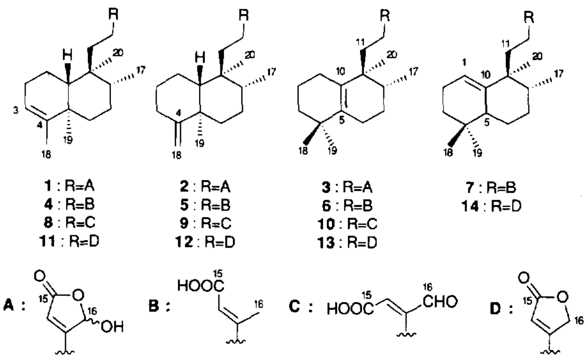
INTRODUCTION

Annonaceae plants are a rich source of bioactive substances [1] and certain genera of this family have recently attracted much interest, since they contain a group of antitumour active tetrahydrofuranic acetogenins [2]. In a continuation of our studies on annonaceous plants, we examined a hexane extract of the stem bark of *Polyalthia longifolia* Thw. *Polyalthia longifolia* is widely distributed in India and commonly known as Asoka in Hindi. Aporphine and azafluorene alkaloids [3, 4], proanthocyanidines [5], and clerodane diterpenes [6–8] have been isolated from the plant. In this paper, we describe the isolation and the structure elucidation of three new clerodanes (**2**, **9** and **12**) and six new *ent*-halimanes [(10 → 9)-friedolabdanes] (**3**, **6**, **7**, **10**, **13** and **14**), along with five known clerodane diterpenes (**1**, **4**, **5**, **8** and **11**).

RESULTS AND DISCUSSION

The stem bark of *P. longifolia* was extracted with hexane. Separation of the extract by silica gel chromatography with hexane–ethyl acetate and ethyl acetate–methanol as eluents afforded three major fractions, A (0.49% based on bark), B (2.3%) and C (0.96%), and one minor fraction, D (0.07%). Further reversed-phase HPLC separation of these fractions furnished **1**–**3** from fraction A, **4**–**7** from fraction B, **8**–**10** from fraction C, and **11**–**14** from fraction D. Compounds **1**–**7** and **11**–**14** were isolated in a pure state, while **8**–**10** were obtained in *ca* 90% purity due to the failure to obtain base line separation of these compounds in HPLC.

Compound **1**, $C_{20}H_{30}O_3$, (EIMS m/z 318 $[M]^+$), was identified as 16-hydroxycleroda-3,13-dien-16,15-olide by comparing the NMR data (Tables 1 and 2) with those reported in the literature [6, 7, 9]. The NMR data further indicated that **1** is a 1:1 epimeric mixture at C-16. This compound was previously isolated from the same species [6, 7], *P. viridis* [9, 10] and *Acritopappus longifolius* [11]. It is interesting to note that the 16*S* configuration was assigned to the compound isolated from the leaves [6] and the stem bark [7] of *P. longifolia*, whereas the 16*R* configuration was assigned to the compound isolated from *P. viridis* [9]. Recently, the compound isolated from *Premna oligotricha* was reported to be a 17:3 mixture of the C-16 epimers [12]. We found that the (*R*)-MTPA esters of the 16*R*- and 16*S*-epimers were readily separable on silica gel TLC, although the C-16 configuration of the separated epimers has not yet been assigned (¹H NMR



1 : R=A
4 : R=B
8 : R=C
11 : R=D

2 : R=A
5 : R=B
9 : R=C
12 : R=D

3 : R=A
6 : R=B
10 : R=C
13 : R=D

7 : R=B
14 : R=D

A : B : C : D :

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Table 1. ^{1}H NMR spectral data of **1**–**14** (500 MHz, $CDCl_3$, TMS as int. standard)

H	**1***	**2***	**3***	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**
1	—	—	—	—	—	—	5.33 (*t*, 3.8)	—	—	—	—	—	—	5.38 (*t*, 3.7)
3	5.18 (*br s*)	—	—	5.19 (*br s*)	—	—	—	5.20 (*br s*)	—	—	5.19 (*br s*)	—	—	—
14	5.83 (*s*)	5.80 (*s*)	5.83 (*s*)	5.69 (*s*)	5.64 (*s*)	5.68 (*s*)	5.67 (*s*)	6.48 (*bs*)	6.46 (*bs*)	6.45 (*bs*)	5.83 (*s*)	5.81 (*s*)	5.83 (*s*)	5.81 (*s*)
16	6.03 (*s*)	6.00 (*m*)	6.00 (*m*)	2.17 (*s*)	2.14 (*s*)	2.17 (*s*)	2.16 (*s*)	9.53 (*s*)	9.53 (*s*)	9.54 (*s*)	4.73 (*s*)	4.72 (*s*)	4.74 (*s*)	4.71 (*s*)
17	0.82 (*d*, 6.0) 0.81† (*d*, 6.0)	0.81 (*d*, 4.0) 0.80† (*d*, 6.0)	0.86 (*d*, 4.0) 0.85† (*d*, 4.0)	0.82 (*d*, 6.4)	0.81 (*d*, 6.4)	0.85 (*d*, 7.3)	0.82 (*d*, 6.4)	0.84 (*d*. 6.8)	0.86 (*d*, 6.4)	0.88 (*d*, 6.8)	0.82 (*d*, 6.4)	0.82 (*d*, 6.2)	0.86 (*d*, 6.3)	0.84 (*d*, 6.4)
18	1.60 (*s*)	4.51 (*s*)	0.97 (*s*)	1.59 (*s*)	4.51 (*s*)	0.96 (*s*)	0.89 (*s*)	1.59 (*s*)	4.51 (*s*)	0.96 (*s*)	1.59 (*s*)	4.51 (*s*)	0.97 (*s*)	0.87 (*s*)
19	1.01 (*s*)	1.05 (*s*)	0.99 (*s*)	1.00 (*s*)	1.05 (*s*)	0.98 (*s*)	0.84 (*s*)	0.99 (*s*)	1.05 (*s*)	0.98 (*s*)	1.01 (*s*)	1.06 (*s*)	0.99 (*s*)	0.85 (*s*)
20	0.78 (*s*)	0.78 (*s*)	0.87 (*s*)	0.74 (*s*)	0.75 (*s*)	0.83 (*s*)	0.91 (*s*)	0.68 (*s*)	0.70 (*s*)	0.81 (*s*)	0.78 (*s*)	0.79 (*s*)	0.87 (*s*)	0.94 (*s*)

*Alcohalic hydrogens of **1**–**3** resonated at δ5.38, 5.39 and 4.80, respectively.
†For the C-16 epimer.

data of the two MTPA esters are shown in the Experimental). 2D NMR studies including ^{1}H–^{1}H COSY, ^{13}C–^{1}H COSY and long range COSY experiments of **1** revealed that the previous ^{13}C assignments [6, 7, 9] need to be revised for several carbons. The corrected ^{13}C assignments are listed in Table 1.

Compound **2** has the same molecular formula (EIMS *m/z* 318 $[M]^+$) as **1**. The ^{1}H NMR spectrum of **2** (Table 1) was similar to that of **1**, except for the presence of the signal of two hydrogens at δ4.51 and the absence of the signals of an olefinic proton at δ5.18 and a vinyl methyl group at δ1.60, observed in the spectra of **1**. This suggested a 4(18)-clerodene (exo-methylene structure) bicyclic structure for **2**. The exo-methylene structure was further supported by the ^{13}C NMR data of **2** in which the signals for C-4 (quaternary) and C-18 (methylene) resonated at δ160.2/160.2 and 102.7/102.8, respectively. The hydroxy-lactone side chain structure was evident from the ^{1}H and ^{13}C NMR data (Tables 1 and 2), as well as the IR absorptions at 3580, 3290, 1755 and 1645 cm^{-1}. On the basis of these data, the structure of **2** was established to be 16-hydroxycleroda-4(18),13-dien-16,15-olide. This compound was a 1:1 epimeric mixture at C-16, which was clearly indicated by the ^{13}C NMR spectral data (Table 2).

Compound **3** had the same molecular formula (EIMS *m/z* 318 $[M]^+$) as **1**. The ^{1}H NMR spectrum of **3** exhibited signals for four (three singlet and one doublet) methyl groups. The ^{13}C NMR spectrum indicated the presence of a tetra-substituted double bond (δ138.3/138.4 and 131.3/131.4) in addition to the side chain double bond. Long range ^{13}C–^{1}H COSY experiments, assisted by the ^{13}C–^{1}H COSY spectrum, revealed the connectivity shown in Fig. 1. Analysis of the ^{13}C–^{1}H COSY spectrum suggested the presence of three allylic methylene carbons (C-1, C-6 and C-12) in the molecule. Further, the ^{13}C NMR data for the bicyclic portion of **1** were in good agreement with those published for dehydroambliol B which has a 5(10)-halimene skeleton [13]. Hence, **3** was established to be 16-hydroxy-*ent*-halima-5(10),13-dien-16,15-olide. The configurations at C-8 and C-9 were presumed to be *R* and *S*, respectively, by analogy with the structures of **1** and **2**, as well as the known *ent*-5(10)-halimene diterpenes of plant origin [14]. Again, **3** was obtained as a 1:1 mixture epimeric at C-16.

Compound **4**, $C_{20}H_{32}O_2$ (EIMS *m/z* 304 $[M]^+$) was identified as kolavenic acid (cleroda-3,13*E*-dien-15-oic acid), a well-known clerodane diterpene, on the basis of spectral comparison (^{1}H, ^{13}C NMR, IR and UV). Isolation of this acid was reported from several plants, e.g. *P. longifolia* [7], *P. viridis* [15], *Hardwickia pinnata* [16] and *Solidago elongata* [17]. Table 2 includes revised ^{13}C NMR assignments for **4** which were obtained by 2D NMR studies.

Compounds **5**–**7** had the same molecular formula as **4**. Compound **5** was shown to have 4(18)-clerodene bicyclic skeleton and 13*E*-unsaturated-15-oic acid side chain by comparing the NMR data with those of **2** and **4** (Tables 1 and 2). Hence, the structure of **5** was established to be cleroda-4(18),13*E*-dien-15-oic-acid. Isolation of this acid as its methyl ester was reported from *Araucaria bidwilli*

Table 2. ^{13}C NMR spectral data of **1**–**14** (125 MHz, $CDCl_3$, TMS as int. standard)*

C	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**
1	18.3	21.3/21.3	25.2/25.2^a	18.3	21.7	25.2^c	120.3	18.1	21.6	25.2^e	18.3	21.7	25.2^g	121.0
2	26.8	32.9	19.9	26.9	33.0	19.4	23.1	26.8	33.2	20.0	26.8	32.9	19.8	23.7
3	120.3/120.4	28.5/28.6	39.8	120.4	28.7	39.9	33.2	120.7	28.6	39.9	120.2	28.6	39.8	33.1
4	144.3/144.3	160.2/160.2	34.5	144.4	160.4	34.5	31.4	144.2	160.7	34.5	144.4	160.1	34.5	31.4
5	38.1	40.0	138.3/138.4	38.2	40.2	137.6	43.5	38.2	40.0	137.2	38.7	40.0	138.6	43.7
6	36.7	37.2	25.7/25.7^a	36.8	37.3	25.8^c	23.6	36.8	37.4	25.0^e	36.7	37.2	25.8^g	23.6
7	27.4	27.4	27.1	27.4	27.4	27.1	29.1	27.6	27.6	27.2	27.3	27.3	27.0	29.0
8	36.3/36.3	36.7/36.7	33.6/33.7	36.3	36.7	33.6	39.2	36.3	37.0	33.5	36.3	36.7	33.7	39.1
9	38.6/38.7	39.2/39.2	40.7	38.8	39.4	40.6	43.0	39.3	40.1	41.1	38.2	39.2	40.7	43.0
10	46.5	48.7	131.1/131.4	46.5	48.7	131.9	141.1	46.6	48.8	132.2	46.5	48.7	131.1	140.6
11	34.8/34.8	34.6/34.7	32.8/32.9	35.0	34.8	34.2	37.2	37.0	36.7	31.6	35.3	35.2	33.3	36.3
12	21.3/21.4	21.7/21.7	22.7/22.7	36.3	36.2	36.2	36.3	19.2	19.2	20.3	22.3	22.3	23.6	23.6
13	171.0	170.9/170.9	170.9/171.0	164.6	164.4	164.8	164.8	157.7	157.7	157.4	171.2	171.1	171.4	171.6
14	116.8	116.7/116.8	116.8/116.9	114.8	114.8	114.7	115.1	133.8	134.2	133.9	115.0	114.9	114.9	114.9
15	172.0	172.1	171.7	171.1	172.0	172.4	172.4	170.9	171.4	170.2	174.0	174.0	174.0	174.1
16	99.3	99.3	99.1/99.1	19.5	19.5	19.9	19.5	194.2	194.1	194.3	73.1	73.0	73.1	73.1
17	15.9	16.0	16.1	15.9	16.0	16.1	15.6	15.9	15.9	16.1	16.0	16.0	16.1	15.6
18	17.9	102.7/102.8	27.6^b	18.0	102.6	27.6^d	28.2	18.0	102.5	27.8^f	17.9	102.8	27.6^h	28.2
19	19.9	20.8/20.8	29.2^b	19.9	20.8	29.2^d	26.0	19.9	20.8	29.2^f	19.9	20.7	29.2^h	25.9
20	18.1	18.0	20.9	18.3	18.1	21.1	22.3	18.0	17.9	21.0	18.1	17.9	20.9	22.2

*Assignment based on INEPT experiments, and particularly in case of **1**, **3**, **7**, **8**, **13** and **14** on ^{13}C–H long range COSY spectra.

$^{a-h}$Assignments may be interchanged.

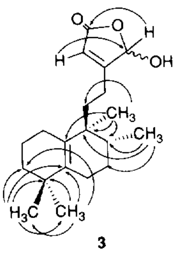

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Fig. 1. Two- and three-bond connectivities revealed by $^{13}C-^{1}H$ long range COSY.

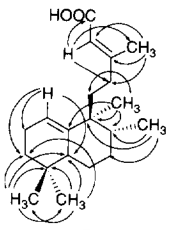

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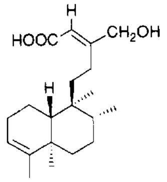
Fig. 2. Two- and three-bond connectivities revealed by $^{13}C-^{1}H$ long range COSY.

[18] and the 13*Z*-isomer of **5** has been reported from *Ageratina ixiocladon* [19].

Compound **6** gave rise to ^1H and ^{13}CNMR data (Tables 1 and 2) which could be explained by combination of 5(10)-*ent*-halimene bicyclic skeleton (e.g. **3**) and 13*E*-unsaturated acid side chain (e.g. **4** and **5**). The structure of **6** was thus established to be *ent*-halima-5(10),13*E*-dien-16-oic acid.

The ^1HNMR spectrum of **7** showed signals of an olefinic proton at δ5.33 and four (three singlet and one doublet) methyl groups, together with the signals due to 13*E*-unsaturated acid side chain at δ5.81 (1H) and 2.17 (3H). The ^{13}CNMR spectrum indicated the presence of a trisubstituted double bond (δ120.3 and 141.1). $^{13}C-^{1}H$ long range COSY experiments revealed the connectivities illustrated in Fig. 2. On the basis of these data, the structure of **7** was determined to be *ent*-halima-1(10), 13*E*-dien-16-oic acid. The α-orientation of H-5, Me-17 and Me-20 groups was confirmed by NOE experiments. Irradiation of the signal at δ1.56 (H-5) resulted in enhancement of the signals at δ0.84 (H-19) and 0.91 (H-20). Compound **7** is the first example of *ent*-1(10)-halimene diterpene lacking an oxygen functionality in the bicyclic portion, although several C-18 oxidized *ent*-1(10)-halimenes are known [20].

Compound **8**, $C_{20}H_{30}O_3$ (EIMS *m/z* 304 $[M]^+$) was identified as 16-oxocleroda-3,13-dien-16-oic acid by comparing the NMR data with those reported previously. However, the confusion regarding the stereochemistry of the 13-ene remained. Phadnis *et al.* [6] and Herz and co-workers [15] have reported the 13*E* structure for the materials (3,13*E*-kolavadien-15-oic acid-16-al) obtained from *P. longifolia* and *P. viridis*, respectively. In contrast, McLaughlin and co-workers reported the isolation of the 13*Z*-isomer from *P. longifolia* (polyalthialdoic acid) [7]. We undertook the following experiments to solve the discrepancy as to the stereochemistry of the 13-ene. Sodium borohydride reduction of **8** afforded the C-16-alcohol **15**. In a NOE experiment irradiation of **15** at δ4.20 (H-16) resulted in enhancement of the signal at δ6.00 (H-14). These results firmly established the 13*E* configuration of **8**. Thus, the structure of **8** was concluded to be 16-oxo-cleroda-3,13*E*-dien-16-oic acid. The most reliable ^{13}CNMR data are shown in Table 2, which are essentially identical with the data of Herz and co-workers (except for the reversed assignment of C-2 and C-11).



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Compounds **9** and **10** have the same molecular formula as **8**. Compound **9** was shown to have a 4(18)-clerodene bicyclic skeleton and a 16-oxo-13*E*-en-15-oic acid side chain by comparing its NMR data with those of **5** and **8** (Tables 1 and 2). Hence, the structure of **9** was determined to be 16-oxocleroda-4(18),13*E*-dien-15-oic acid. Similarly, **10** was determined to be 16-oxo-*ent*-halima-5(10),13*E*-dien-15-oic acid on the basis of a NMR comparison with **6** and **8**.

Compounds **11**–**14** have the same molecular formula, $C_{20}H_{30}O_2$ (EIMS *m/z* 302 $[M]^+$). Compound **11** was identified as solidagolactone I(cleroda-3,13*Z*-dien-16,15-olide) by comparing its ^1HNMR data with those reported [20]. The ^{13}CNMR data listed in Table 2 are in accord with the structure. The α-orientation of the 17-, 19- and 20-methyl groups was confirmed by NOE experiments on **11**.

Compound **12** was shown to have a 4(18)-clerodene bicyclic skeleton and an unsaturated γ-lactone structure (e.g. **11**) by comparing its NMR data (Tables 1 and 2) with those of **5** and **11**. Hence, the structure of **12** was established to be cleroda-4(18),13-dien-16,15-olide. Compound **13** was established to be *ent*-halima-5(10),13-dien-16,15-olide by comparing the NMR data with those of **6** and **11** (Tables 1 and 2). Compound **14** was similarly established to be *ent*-halima-1(10),13-dien-16,15-olide by NMR comparison.

In conclusion, we have isolated and characterized nine new bicyclic diterpenes, including 1(10)- and 5(10)-*ent*-halimanes from the stem bark of *P. longifolia*. This is the first report of the isolation of *ent*-halimane diterpenes from Annonaceae plants.

EXPERIMENTAL

Mps: uncorr.; ^1H and ^{13}CNMR: 500 and 125 MHz, respectively; IR and UV; $CHCl_3$ and MeOH, respectively.

Isolation. The stem bark of *P. longifolia* (1 kg), collected from Varanasi, India in July 1990, was powdered and extracted with hexane for 2 days in a Soxhlet extractor. The petrol extract (50 g) was chromatographed over a column of silica gel (500 g) with hexane–EtOAc and then with EtOAc–MeOH as an eluent. Elution with hexane–EtOAc (10:1) afforded fr. B (4.9 g), and elution with hexane–EtOAc (6:1) afforded fr. A (23.3 g). Continued elution with EtOAc–MeOH (10:1) gave fr. C (9.6 g). Rechromatography of the frs eluted before fr. B with hexane–EtOAc (12:1) gave fr. D (0.7 g).

HPLC sepn (column, STR PREP-ODS, 25 × 2 cm i.d.; solvent, MeOH–H_2O (7:1); flow rate, 8 ml min^{-1}; detected by UV at 235 nm) of a part of fr. B (280 mg) gave a mixt. of **2** and **3** (R_t 22.5 min, 60 mg), and **1** (R_t 21.0 min, 50 mg). The mixt. of **2** and **3** was further sepd under the same conditions except for the use of MeCN–H_2O (20:1) as a solvent to furnish **2** (R_t 11.2 min, 24 mg) and **3** (R_t 12.3 min, 27 mg). A part of fr. A (680 mg) was sepd similarly with MeOH–H_2O–HOAc (180:120:1) as solvent to furnish **6** (R_t 35.8 min, 12 mg), **5** (R_t 36.8 min, 169 mg), **7** (R_t 37.8 min, 15 mg), and **4** (R_t 41.1 min, 204 mg). A part of fr. C (210 mg) was similarly chromatographed with MeOH–H_2O–HOAc (200:20:1) to afford partially purified **9** (R_t 31.3 min), **10** (R_t 32.7 min) and **8** (R_t 35.0 min). Repeated HPLC sepn furnished samples (with *ca* 90% purity) of **9** (8 mg), **10** (14 mg) and **8** (20 mg). A part of fr. D (181 mg) was further sepd with MeCN–H_2O (20:1) as solvent to give **12** (R_t 22.8 min, 22 mg), **11** (R_t 23.9 min, 88 mg), **13** (R_t 26.4 min, 35 mg) and **14** (R_t 27.6 min, 8 mg). Yields based on the bark ranged from 0.003% for **14** to 0.4% for **1**.

16-*Hydroxycleroda*-3,13-*dien*-16,15-*olide* (**1**). Amorphous solid; $[\alpha]_D^{25}$ −27° ($CHCl_3$; *c* 0.68) (lit. −70.58° [6], −48.7° [9], −42° [11]); IR ν_{max} cm^{-1}: 3590, 3300, 2970, 2943, 1760, 1652; UV λ_{max}^{MeOH} (log ε): 211 nm (4.1); EIMS *m/z* (rel. int.): 318 $[M]^+$ (16), 303 (6), 300 (6), 285 (16), 191 (72), 135 (36), 107 (100), 95 (91), 55 (86), 41 (96). Compound **1a**: the less polar (*R*)-MTPA ester of **1**, R_f 0.31 on Merck F_{254} silica plate (solvent hexane–EtOAc 10:1), oil ^1H NMR: δ0.64 (*d*, *J* = 6.8 Hz, H_3-17), 0.72 (*s*, H_3-20), 0.97 (*s*, H_3-19), 1.61 (*s*, H_3-18), 5.21 (*br s*, H-3), 5.94 (*s*, H-14), 6.97 (*s*, H-16). Compound **1b**: the more polar (*R*)-MTPA ester of **1**, R_f 0.25 under the same conditions described for **1a**, oil, ^1H NMR: δ0.73 (*s*, H_3-20), 0.76 (*d*, *J* = 6.8 Hz, H_3-17), 0.98 (*s*, H_3-19), 1.61 (*s*, H_3-18), 5.17 (*br s*, H-3), 5.95 (*s*, H-14), 6.99 (*s*, H-16).

16-*Hydroxycleroda*-4(18),13-*dien*-16,15-*olide* (**2**). Oil; $[\alpha]_D^{25}$ +10° ($CHCl_3$; *c* 1.20); IR ν_{max} cm^{-1}: 3580, 3290, 2930, 1755, 1645, 893; UV λ_{max}^{MeOH} (log ε): 211 nm (4.1); EIMS *m/z* (rel. int.): 318 $[M]^+$ (8), 303 (4), 300 (5), 285 (8), 191 (73), 135 (49), 107 (79), 95 (100), 55 (64), 41 (82); Found: C, 75.13; H, 9.68. $C_{20}H_{30}O_3$ requires C, 75.43; H, 9.49%.

16-*Hydroxy*-ent-*halima*-5(10),13-*dien*-16,15-*olide* (**3**). Oil; $[\alpha]_D^{25}$ + 21°($CHCl_3$; *c* 0.54); IR ν_{max} cm^{-1}: 3575, 3300, 2960, 2925, 1755, 1645; UV λ_{max}^{MeOH} (log ε): 211 nm (4.1); EIMS *m/z* (rel. int.): 318 $[M]^+$ (3), 285 (1), 191 (100), 135 (11), 95 (12), 55 (13), 41 (15).

Cleroda-3,13E-*dien*-15-*oic acid* (**4**). Oil; $[\alpha]_D^{25}$ −44° ($CHCl_3$; *c* 0.68); IR ν_{max} cm^{-1}: 3000 (br), 2960, 1690, 1637, 1253; UV λ_{max}^{MeOH} (log ε): 218 nm (4.1); EIMS *m/z* (rel. int.): 304 $[M]^+$ (32), 289 (4), 271 (7), 191 (58), 135 (35), 107 (88), 95 (100), 55 (70), 41 (99).

Cleroda-4(18),13E-*dien*-15-*oic acid* (**5**). Crystals, mp 106–108° (from MeOH–H_2O); $[\alpha]_D^{25}$ + 5° ($CHCl_3$; *c* 0.627); IR ν_{max} 3000 (*br*), 2930, 1690, 1637, 1253, 893; UV λ_{max}^{MeOH} (log ε): 218 nm (4.1); EIMS *m/z* (rel. int.): 304 $[M]^+$ (19), 289 (6), 286 (4), 271 (6), 191 (99), 135 (44), 95 (100), 81 (56), 55 (64), 41 (96); Found: C, 78.68; H, 10.63. $C_{20}H_{32}O_2$ requires C, 78.90 H, 10.59%.

Ent-*halima*-5(10),13E-*dien*-15-*oic acid* (**6**). Crystals, mp 123–124° (from MeOH–H_2O); $[\alpha]_D^{25}$ +25.8° (MeOH; *c* 0.62); IR ν_{max} cm^{-1}: 3000 (*br*), 2940, 1690, 1637, 1253; UV λ_{max}^{MeOH} (log ε): 218 nm (4.1); EIMS *m/z* (rel. int.): 304 $[M]^+$ (2), 289 (1), 271 (1), 244 (2), 191 (100), 135 (19), 95 (19), 91 (18), 55 (18), 41 (29).

Ent-*halima*-1(10),13E-*dien*-15-*oic acid* (**7**). Crystals, mp 111–112° (from MeOH–H_2O); $[\alpha]_D^{25}$ + 67.5° (MeOH; *c* 0.37); IR ν_{max} cm^{-1}: 3000 (*br*), 2940, 1690, 1637, 1255; UV λ_{max}^{MeOH} (log ε): 218 nm (4.1); EIMS *m/z* (rel. int.): 304 $[M]^+$ (3), 289 (5), 271 (1), 244 (3), 191 (100), 135 (33), 95 (21), 55 (24), 41 (39).

16-*Oxocleroda*-3-13E-*dien*-15-*oic acid* (**8**). Oil; $[\alpha]_D^{25}$ − 88.8° (MeOH; *c* 0.5) (lit. − 78.6° [15], − 36.7° [7]); IR ν_{max} cm^{-1}; 3000 (*br*), 2965, 2935, 1703, 1644; UV λ_{max}^{MeOH} (log ε): 235 nm (3.9); EIMS *m/z*: 318 $[M]^+$.

16-*Oxocleroda*-4(18),13E-*dien*-15-*oic acid* (**9**). Oil; IR ν_{max} cm^{-1}: 3000 (*br*), 2965, 2935, 1703, 1644, 895; UV λ_{max}^{MeOH} (log ε): 230 nm (3.8); EIMS *m/z*: 318 $[M]^+$.

16-*Oxo*-ent-*halima*-5(10),13E-*dien*-15-*oic acid* (**10**). Oil; IR ν_{max} cm^{-1}: 3000 (*br*), 2965, 2935, 1703, 1644; UV λ_{max}^{MeOH} (log ε): 235 nm (4.0); EIMS *m/z*: 318 $[M]^+$.

16-*Hydroxycleroda*-3,13E-*dien*-15-*oic acid* (**15**). Treatment of acid **8** (20 mg) with $NaBH_4$ (10 mg) in MeOH (1 ml) at room temp. for 1 hr followed by usual work-up and purification over silica gel gave **15** (13 mg, 61%) as an oil. $[\alpha]_D^{25}$ −60.0° (MeOH; *c* 0.50) (lit. −67.9° [6]); ^1H NMR: δ0.71 (*s*, H-20), 0.84 (*d*, *J* = 6.4 Hz, H-17), 0.99 (*s*, H-19), 1.58 (*s*, H-18), 2.38 (*m*, H-12), 4.20 (*s*, H-16), 5.20 (*s*, H-3), 6.00 (*s*, H-14). HREIMS Found 320.2333 $[M]^+$, $C_{20}H_{32}O_3$ requires 320.2351.

Cleroda-3,13-*dien*-16,15-*olide* (**11**). Crystals; $[\alpha]_D^{25}$ −52.5° (MeOH; *c* 1.2); IR ν_{max} cm^{-1}: 3025, 2970, 2940, 1784, 1747, 1641. UV λ_{max}^{MeOH} (log ε): 218 nm (3.9); EIMS *m/z* (rel. int.): 302 $[M]^+$ (32), 289 (4), 271 (7), 191 (58), 135 (35), 107 (88), 95 (100), 55 (70), 41 (99). HRFABMS Found 303.2343 $[MH]^+$, $C_{20}H_{31}O_2$ requires 303.2324.

Cleroda-4(18),13-*dien*-16,15-*olide* (**12**). Crystals, mp 66–67° (MeOH–H_2O); $[\alpha]_D^{25}$ + 15.2° (MeOH; *c* 1.9); IR ν_{max} cm^{-1}: 2940, 1783, 1742, 1638, 895; UV λ_{max}^{MeOH} (log ε): 218 nm (3.9); EIMS *m/z* (rel. int.): 302 $[M]^+$ (14), 287 (23), 191 (100), 135 (73), 107 (40), 95 (69), 55 (33), 41 (36); Found: C, 78.68; H, 10.63. $C_{20}H_{32}O_2$ requires C 78.90, H 10.59%.

Ent-*halima*-5(10),13-*dien*-16,15-*olide* (**13**). Amorphous; $[\alpha]_D^{25}$ + 18.7° (MeOH; *c* 1.3); IR ν_{max} cm^{-1}: 3025, 2940, 2873, 1690, 1637, 1253; UV λ_{max}^{MeOH} (log ε): 218 nm (4.1); EIMS *m/z* (rel. int.): 302 $[M]^+$ (12), 287 (12), 191 (100).

HRFABMS Found 303.2323 $[MH]^+$, $C_{20}H_{31}O_2$ requires 303.2324.

Ent-*halima*-1(10),13-*dien*-16,15-olide (**14**). Amorphous; $[\alpha]_D^{25}$ + 77.8° (MeOH; *c* 1.2); IR ν_{max} cm^{-1}: 3025, 2940, 1783, 1745, 1638; UV λ_{max}^{MeOH} (log ε): 218 nm (4.1); EIMS *m/z* (rel. int.): 302 $[M]^+$ (21), 287 (23), 191 (100), 135 (48), 107 (22), 95 (17), 55 (12), 41 (13). HRFABMS Found 303.2320 $[MH]^+$, $C_{20}H_{31}O_2$ requires 303.2324.

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REFERENCES

1. Leboeuf, M., Cave, A., Bhaumik, P. K., Mukherjee, B. and Mukherjee, R. (1982) *Phytochemistry* **21**, 2783.
2. Rupprecht, J. K., Hui, Y.-H. and McLaughlin, J. L. (1990) *J. Nat. Prod.* **53**, 237.
3. Jossang, A., Leboeuf, M. and Cave, A. (1982) *Tetrahedron Letters* **23**, 5147.
4. Wu, Y.-C., Duh, C.-Y., Wang, S.-K., Chen, K.-S. and Yang, T.-H. (1990) *J. Nat. Prod.* **53**, 1327.
5. Wu, Y.-C. (1989) *Heterocycles* **29**, 463.
6. Phadnis, A. P., Patwardhan, S. A., Dhaneshwar, N. N., Tavale, S. S. and Row, T. N. G. (1988) *Phytochemistry* **27**, 2899.
7. Zhao, G., Jung, J. H., Smith, D. L., Wood, K. V. and Mclaughlin, J. L. (1991) *Planta Med.* **57**, 380.
8. Chakrabarty, M. and Nath, A. C. (1992) *J. Nat. Prod.* **55**, 256.
9. Kijjoa, A., Pinto, M. M. M. and Herz, W. (1989) *Planta Med.* **55**, 205.
10. Kijjoa, A., Pinto, M. M. M., Pinho, P. M. M., Tantisewie, B. and Herz, W. (1990) *Phytochemistry* **29**, 653.
11. Bohlmann, F., Ahmed, M., Jakupovic, J., King, R. M. and Robinson, H. (1984) *Rev. Latinamer. Quim.* **15**, 16.
12. Habtemariam, S., Gary, A. I. and Waterman, P. G. (1992) *Planta Med.* **58**, 109.
13. Walker, R. P. and Faulkner, D. J. (1981) *J. Org. Chem.* **46**, 1098.
14. Pascual Teresa, J. de, Urones, J. G., Marcos, I. S., Bermejo, F. and Basabe, P. (1983) *Phytochemistry* **22**, 2783.
15. Kijjoa, A., Pinto, M. M. M., Pinho, P. M. M. and Herz, W. (1993) *Phytochemistry* **34**, 457.
16. Misra, R., Pandey, R. C. and Dev, S. (1964) *Tetrahedron Letters* **49**, 3751.
17. Anthonsen, T. and McCrindle, R. (1969) *Acta Chem. Scand.* **23**, 1068.
18. Caputo, R. and Mangoni, L. (1974) *Phytochemistry* **13**, 467.
19. Tamayo-Castillo, G., Jakupovic, J., Bohlmann, F., Castro, V. and King, R. M. (1989) *Phytochemistry* **28**, 139.
20. Pascual Teresa, J. de, Urones, J. G. and Carrillo, H. (1978) *An. Quim.* **74**, 489.
21. Okazaki, T., Ohsuka, A. and Kotake, M. (1973) *Nippon Kagaku Kaishi* 584.
22. Urones, J. G., Pascual Teresa, J. de, Marcos, I. S., Martin, D. D., Garrido, N. M. and Guerra, R. A. (1987) *Phytochemistry* **26**, 1077.