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Four New Bioactive Lobane Diterpenes of the Soft Coral *Lobophytum pauciflorum* from Mindoro, Philippines

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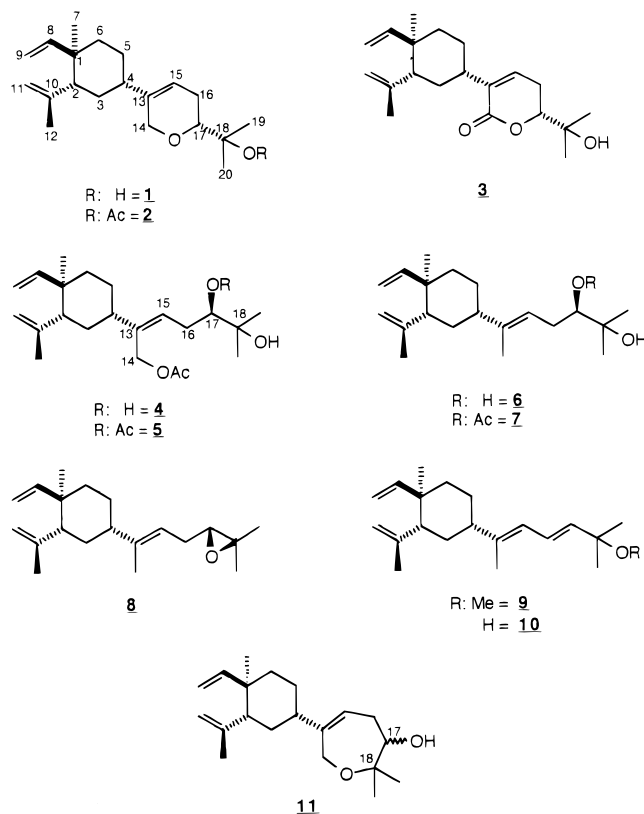
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The marine soft coral *Lobophytum pauciflorum* collected from Mindoro Island, Philippines, yielded four new lobane diterpene derivatives: the acetate congeners of epoxylobatrienol and lobatrienediol (**2** and **7**, respectively), a methoxyl congener of lobatetraene (**10**), and an oxepin congener of lobatrienetriol (**11**), and six known derivatives (**1**, **3–6**, and **8**). The structures of the new compounds were unambiguously established on the basis of NMR spectroscopic (¹H, ¹³C, COSY, ¹H-detected direct, and long-range ¹³C–¹H correlations) and mass spectrometric (EIMS) data. All of the compounds were active against the phytopathogenic fungus *Cladosporium cucumerinum*. Compound **1** was found to be active against the Gram-positive bacteria *Bacillus subtilis* and the yeast *Saccharomyces cerevisiae*. The isolated lobane diterpenes were also active in the brine shrimp lethality test. In the latter bioassay, compounds **8** and **10** were the most active congeners with LC₅₀'s of 0.64 and 4.18 μg/mL, respectively.

Alcyonarians are known as rich sources of sesquiterpenes and diterpenes with unique structural diversity and pronounced chemical activities. The lobane diterpenes are closely related to the known β-elemene (**12**), which is a sesquiterpene congener of germacrene (**13**), with an additional acyclic or cyclic isoprenyl unit. Lobanes have been isolated from a gorgonian species *Eunicea fusca*¹ and from two different genera of marine soft corals, *Sinularia*^{2–4} and *Lobophytum*.⁵ The soft coral species *Lobophytum pauciflorum* of the Andaman and Nicobar coasts of India^{6–8} has afforded lobane derivatives similar to those of an unidentified *Lobophytum* species collected at the Great Barrier Reef, Australia.⁵ Specimens of *L. pauciflorum* collected off the Mindoro Island, Philippines, showed the presence of four new lobane congeners. In the present paper, we describe the isolation and structure elucidation of the four new lobanes (**2**, **7**, **10**, and **11**) and six other known derivatives (**1**, **3–6**, and **8**) and report on their antibacterial, fungicidal, and cytotoxic properties.

Results and Discussion

The organo-solvent extraction of the soft coral *L. pauciflorum* Ehrenberg (Family Alcyoniidae) gave a crude extract that on Si gel chromatography afforded 10 compounds. The identities of the known lobane derivatives (**1**, **3–6**, and **8**) were established by comparison with published data.^{1,3–8} Two-dimensional homonuclear correlation spectroscopy (¹H COSY) and



heteronuclear ¹H-detected ¹³C multiple quantum coherence (HMBC, HMQC) spectra afforded independent, unambiguous confirmation of the signal assignments, substituent position, and total structure of the new compounds (Tables 1 and 2). A careful inspection of the ¹³C and ¹H NMR spectra of the isolated compounds showed resonances and coupling constants identical to

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Table 1. ^{13}C NMR data of compounds **1**, **2**, **6**, **7**, and **9–11** in CDCl_3

C no.	1	2	6	7	9^a	10	11
1	39.8 s	39.7 s	39.8 s	39.7 s	39.6 s	39.8 s	39.9 s
2	52.7 d	52.8 d	52.8 d	52.7 d	52.3 d	52.8 d	52.8 d
3	32.8 t	32.8 t	33.0 t	32.8 t	32.5 t	32.7 t	33.0 t
4	41.7 d	41.7 d	47.7 d	47.6 d	47.5 d	47.7 d	44.7 d
5	24.8 t	25.0 t	26.8 t	26.7 t	26.4 t	26.6 t	27.1 t
6	39.9 t	39.9 t	39.9 t	39.9 t	39.7 t	39.9 t	39.7 t
7	16.6 q	16.6 q	16.7 q	16.5 q	16.4 q	16.6 q	16.7 q
8	150.1 d	150.1 d	150.3 d	150.3 d	150.1 d	150.1 d	150.1 d
9	112.2 t	112.3 t	112.1 t	111.8 t	109.6 t	109.9 t	110.0 t
10	147.5 s	147.5 s	147.7 s	147.7 s	147.5 s	147.7 s	147.4 s
11	110.0 t	110.0 t	109.9 t	109.8 t	112.0 t	112.2 t	112.3 t
12	23.7 q	24.8 q	24.8 q	25.2 q	24.7 q	24.8 q	24.8 q
13	141.2 s	141.4 s	143.8 s	142.4 s	143.2 s	143.2 s	147.3 s
14	68.1 t	68.4 t	23.7 q	24.8 q	15.3 q	15.4 q	64.4 t
15	116.4 d	116.3 d	118.9 d	118.4 d	122.9 d	122.6 d	117.8 d
16	27.0 t	27.1 t	30.4 t	28.2 t	123.0 d	126.0 d	31.8 t
17	80.3 d	78.3 d	77.9 d	79.6 d	139.1 d	136.8 d	78.2 d
18	71.7 s	83.1 s	72.7 s	72.3 s	70.9 s	75.1 s	75.9 s
19	26.2 q	22.5 q	26.2 q	26.8 q	29.8 q	26.1 q	22.5 q
20	25.3 q	22.6 q	26.2 q	26.8 q	29.8 q	26.0 q	25.3 q
21						50.5 q	
OAc		170.6 s		170.8 s			
OAcMe		21.7 q		21.0 q			

^a Taken from ref 1.

those of β -elemene, which established the presence of the elemene ring system.

Compounds **2** and **7** were obtained as yellow oils. They showed molecular ion peaks $[\text{M}]^+$ at m/z 346 and 348 in the EIMS that are compatible with the molecular composition of $\text{C}_{22}\text{H}_{34}\text{O}_3$ and $\text{C}_{22}\text{H}_{36}\text{O}_3$. Compounds **2** and **7** are the acetate congeners of **1** and **6**, which were confirmed by comparison of their ^{13}C and ^1H NMR data. In each case, the ^{13}C NMR spectrum showed an additional signal of a carbonyl group and methyl group, indicating the presence of the acetyl substituent. Characteristic changes in chemical shifts of neighboring carbons were observed. These data confirmed the acetylation of the appropriate hydroxyl group and established **2** as 14,17-epoxyloba-8,10,13(15)-trien-18-ol 18-acetate and **7** as loba-8,10,13(15)-triene-17,18-diol 18-acetate.

Compound **10** was obtained as a colorless oil and showed a molecular ion peak $[\text{M}]^+$ at m/z 302 in the EIMS, which is compatible with the molecular composition of $\text{C}_{21}\text{H}_{34}\text{O}$. The ^{13}C and ^1H NMR data of **10** were comparable to those of lobatetraene (**9**),¹ a lobane congener isolated from the gorgonian species *Eunicea fusca*. A difference of 14 mass units in the mass

spectrum of **10** compared to that of **9** suggests a possible methylation of the hydroxyl function at C-18. In the ^{13}C NMR spectrum of **10**, differences in the spectrum of **9** were observed with changes in chemical shifts for C-16, C-17, C-18, C-19, and C-20, and an additional methoxyl signal was present at δ 50.5. These changes were only compatible with methylation of the hydroxyl function at C-18 of **9**. Confirmation of this was afforded by a HMBC spectrum that unambiguously established the atom connectivities in **10**. In this spectrum, a CH direct correlation was observed between the singlet at δ 3.15 and the methyl carbon at δ 50.5. Long-range correlations were found between the methoxyl protons at δ 3.15 and the quarternary carbon at δ 75.1 (C-18) and from the olefinic proton at δ 6.38 (H-16) to C-18. Thus, **10** was 18-methoxyloba-8,10,13(15),16(17)-tetraene.

Compound **11** was obtained as a colorless oil and showed the molecular ion peak $[\text{M}]^+$ at m/z 304 in the EIMS, which is compatible with the molecular composition of $\text{C}_{20}\text{H}_{32}\text{O}_2$. Compound **11** is an oxepin congener of **1**. Its structure was confirmed by comparison of the ^{13}C and ^1H NMR data with those of **1**. Changes in chemical shifts in the ^{13}C and ^1H NMR spectrum were observed for the additional cyclic isoprenyl unit, particularly for C-13, C-14, C-16, and C-18 with changes in shifts of 4–6 ppm. The HMBC spectrum afforded the unambiguous confirmation of the oxepin ring system as shown by a long-range correlation of H-14 to C-18, indicating the cyclization of the hydroxyl at C-14 with that of C-18 in contrast to that of C-17 as in **1**. Thus, the structure of **11** was 14,18-epoxyloba-8,10,13(15)-trien-17-ol.

The relative and absolute configurations of the various asymmetric centers in the molecule have been established for the known compounds (**1**, **3–6**, and **8**),^{3,4} and those of the four new compounds must be the same from the comparison of the NMR data in Tables 1 and 2. The similarity in ^{13}C shifts of C-1 to C-12 in all compounds and the axial orientation of H-2 with couplings of 5.3 and 10.7 Hz indicates the relative stereochemistry of the cyclohexane system is maintained throughout. Similarly, the axial orientation of H-17 with coupling of 3.4 and 10.8 Hz is maintained in **2**, and the ^{13}C shifts imply the configuration at the double bonds in the side chains of **7** and **10** are the same as **6** and **9**, respectively. The relative stereochemistry of

Table 2. ^1H NMR Data^a of Compounds **1**, **2**, **6**, **7**, and **9–11** in CDCl_3

H no.	1	2	6	7	9^b	10	11
3	1.51 m	1.52 m	1.47 m	1.47 m	1.61 m	1.60 m	1.52 m
14	4.20 bs	4.17 bs	1.65 s	1.62 s	1.78 s	1.78 s	A 4.18 dd (16.3, 1.6) B 4.10 bd (16.4)
15	5.57 dp (5.6, 1.7)	5.55 bd (5.6)	5.26 bt (7.0)	5.14 bdd (7.0, 8.0)	5.88 bd (10.6)	5.88 bd (11.5)	5.36 t (5.2)
16A	2.15 m	2.12 m	2.18 m	2.32 m	6.44 dd (10.7, 15.5)	6.38 dd (10.8, 15.5)	2.50 m
16B	1.95 m	1.95 m					
17	3.26 dd (3.5, 10.8)	3.73 dd (3.4, 10.8)	3.36 dd (4.7, 8.3)	4.83 dd (4.0, 9.4)	5.60 d (5.5)	5.57 d (15.5)	3.64 dd (7.7, 8.0)
19	1.17 s	1.45 s	1.17 s	1.21 s	1.35 s	1.28 s	1.25 s
20	1.21 s	1.49 s	1.22 s	1.21 s	1.42 s	1.28 s	1.27 s
21						3.15 s	
OAcMe		1.98 s		2.04 s			

^a Chemical shifts are given in ppm relative to internal TMS and coupling constants in Hz are in parentheses. The ^1H data for systems containing carbons 1–12 are essentially identical, except for H-3, in all compounds, and hence, only the data for **1** are given here. **1**: δ 1.98 (dd, H-2, $J(2-3) = 5.3, 10.7$ Hz), 1.83 (m, H-5A), 1.65–1.37 (m, H-6A, H-4), 1.51 (m, H-5B), 1.47 (m, H-6B), 0.99 (bs, H-7), 5.81 (dd, H-8, $J(8-9A) = 17.4$ Hz, $J(8-9B) = 10.2$ Hz), 4.91 (bd, H-9A/B), 4.82 (p, H-11A, $J(11A-12) = 1.8$ Hz, $J(11A-11B) = 1.8$ Hz), 4.57 (bs, H-11B), 1.70 (bs, H-12). In the 2D COSY spectrum, long-range couplings are observed between the methyl group H-7 and H-8, H-11A, H-2 and H-6B, and between H-2 and H-11B. ^b Taken from ref 1.

Table 3. Bioactivities of the Compounds Isolated from *Lobophytum pauciflorum*

compd no.	zone of inhibition fungicidal activity against <i>C. cucumerinum</i> (mm diameter) ^a				brine shrimp lethality test <i>A. salina</i> ($\mu\text{g/mL}$)
	0.80	0.40	0.20	0.10	($\text{LC}_{50} \pm \text{SE}$)
1	15.0	14.0	10.0	5.0	19.80 ± 0.66
2	not tested	10.0	8.0	6.0	33.96 ± 0.14
3	15.0	13.0	9.0	6.0	not tested
4	30.0	20.0	10.0	5.0	59.02 ± 0.18
5	15.0	13.0	10.0	5.0	60.40 ± 0.17
6	17.0	14.0	9.0	6.0	10.50 ± 0.85
7	13.0	9.0	5.0	2.0	19.10 ± 0.35
8	5.0	0.0	0.0	0.0	0.64 ± 0.63
10	2.0	0.0	0.0	0.0	4.18 ± 0.32
11	17.0	14.0	10.0	5.0	not tested
mimosamycin		18.0	15.0		

^a Dose: μmol .

C-17 in **11** does not follow from the present data. The optical rotation values for the known compounds were similar to those previously reported.^{1,3-8}

All compounds isolated were analyzed for antibacterial and fungicidal activity. The compounds were tested for antibacterial activity against *Bacillus subtilis*, *Staphylococcus aureus*, and *Escherichia coli* and also against the yeast *Saccharomyces cerevisiae*. Only compound **1** was found to be active against the Gram-positive *B. subtilis* and the yeast *S. cerevisiae*, causing an inhibition zone of 8 and 7 mm in diameter, respectively. No inhibition, however, was observed for *S. aureus* and *E. coli*.

All compounds were active against the phytopathogenic fungus *C. cucumerinum*. The lobatrienetriol congeners (**4** and **5**) were most active, followed by the oxepin congener (**11**) and then by the lobatrienediol congeners (**6** and **7**) and the oxinine congeners (**1**–**3**), while the epoxide congener (**8**) and the diene congener (**10**) showed the weakest activity (Table 3).

All of the isolated compounds were also tested in the brine shrimp lethality assay except for **3** and **11** due to their very low yield. For this assay, the epoxide congener (**8**) was found to be the most active compound with an LC_{50} of $0.64 \mu\text{g/mL}$, followed by the diene derivative (**10**) with a LC_{50} of $4.18 \mu\text{g/mL}$, and then by the lobatrienediol congeners (**6** and **7**) and by the oxinine congeners (**1**–**3**). The lobatrienetriol congeners (**4** and **5**) showed very weak activity with LC_{50} 's of 59.02 and $60.40 \mu\text{g/mL}$, respectively (Table 3).

It is remarkable that the observed antibacterial activity of **1**, the fungicidal activity of the lobatrienetriol congeners (**4** and **5**), and the brine shrimp lethality of the epoxide congener (**8**) and the diene congener (**10**) are obviously not caused by general toxicity but are rather due to different modes of action and specific target requirements, which are apparently strongly influenced by the chemical structure of the studied compounds. This specificity in the mode of action of this series of lobane derivatives seems to be differentiated by the additional isoprenyl unit attached to the basic β -elemene ring. Taking compound **1** as the reference compound, the opening of the heteroring weakens its lethality in the brine shrimp test and strengthens the fungicidal activity, as with the lobatrienetriol congeners, while the dehydroxylation of the lobatrienetriol conge-

ner at C-14 to its diol derivative results in an increase in brine shrimp lethality and decrease of fungicidal activity, as in compounds **6** and **7**. The formation of an epoxide bridge in a lobatrienediol derivative or an increase in unsaturation of the chain system causes a large increase in the brine shrimp lethality and abrupt loss of fungicidal activity. However, it has been generally observed that acetylation of the hydroxyl substituent results in a weakening of the activity in all the bioassays performed.

Experimental Section

General Experimental Procedures. ^1H and ^{13}C NMR spectra (chemical shifts in ppm) were recorded on Bruker ARX 400 NMR and AVANCE DMX 600 NMR spectrometers. Mass spectra (EIMS) were measured on a Finnigan MAT 8430 mass spectrometer. Optical rotations were determined on a Perkin-Elmer-241 MC polarimeter. UV spectra were recorded in hexane.

Solvents were distilled prior to use, and spectral grade solvents were used for spectroscopic measurements. TLC was performed on precoated TLC plates with Si gel 60 F254 (Merck, Darmstadt, Germany). The compounds were detected by spraying the TLC plates with anisaldehyde reagent.

Animal Material. Specimens of the soft coral *L. pauciflorum* were collected by snorkelling off the shores of Mindoro Island, Philippines, in April 1994. The samples were frozen immediately and then freeze-dried prior to transport to the University of Würzburg, Germany. A voucher fragment is kept in 70% ethanol under the voucher no. RE-11.04.94 in the Nationaal Natuurhistorisch Museum, Leiden.

Extraction and Isolation. The freeze-dried samples of *L. pauciflorum* (30 g) were extracted with acetone and MeOH (300 mL \times 2 for each) successively. The total extract was evaporated under reduced pressure and was partitioned between EtOAc (50 mL \times 5) and H₂O (50 mL). The organic fraction was taken to dryness (2.5 g) and chromatographed over a Si gel column (mobile phase $\text{CH}_2\text{Cl}_2/\text{MeOH}$ 98:2), and 18 fractions were obtained. The nonpolar fractions 3, 4 and 5 afforded **10** (11.1 mg, 0.037%), **8** (39.3 mg, 0.131%), and **2** (3.1 mg, 0.010%), respectively. The semipolar fractions 11, 12, and 13 contained the major compound **1** (171.2 mg, 0.571%). Compounds **3** (2.1 mg, 0.007%), **7** (15.6 mg, 0.052%), and **11** (2.8 mg, 0.009%) were obtained from the polar fraction 14. The more polar fractions 15 and 16 contained **4** (18.4 mg, 0.061%), **5** (15.0 mg, 0.050%), and **6** (42.0 mg, 0.140%). The pure compounds were obtained by rechromatography on Si 60 Lobar columns using different ratios of hexane/EtOAc as eluent, 95:5 for the nonpolar fractions 3–5; 90:10 for the semipolar fractions 11–13, 70:30 for the polar fraction 14, and 60:40 for the more polar fractions 15 and 16.

14,17-Epoxyloba-8,10,13(15)-triene-18-ol 18-acetate (2) was obtained as a yellow viscous oil: UV λ_{max} (hexane) 199 (ϵ 4300); $[\alpha]_{\text{D}}^{25} +47.31^\circ$ (c 0.31, CHCl_3) ($\text{C}_{22}\text{H}_{34}\text{O}_3$); EIMS (70 eV) m/z $[\text{M}]^+$ 346 (6), 318 (6), 300 (30), 285 (60), 243 (36), 229 (24), 201 (22), 187 (26), 161 (34), 147 (32), 133 (30), 121 (42), 107 (44), 93 (44), 81 (62), 71 (52), 55 (40), 43 (100).

Loba-8,10,13(15)-triene-17,18-diol 18-acetate (7) was obtained as a yellow viscous oil: UV λ_{max} (hexane)

199 (ϵ 4350); $[\alpha]_D +52.7^\circ$ (c 1.56, CHCl_3) ($\text{C}_{22}\text{H}_{36}\text{O}_3$); EIMS (70 eV) m/z $[\text{M}]^+$ 348 (2), 333 (4), 288 (40), 270 (24), 230 (22), 216 (24), 189 (38), 161 (40), 147 (40), 133 (40), 125 (52), 109 (56), 93 (48), 81 (52), 59 (48), 43 (100).

18-Methoxyloba-8,10,13(15),16(17)-tetraene (10) was obtained as a colorless viscous oil: UV λ_{max} (hexane) 199 (ϵ 8690), 240 (ϵ 1710); $[\alpha]_D -2.6^\circ$ (c 0.33, CHCl_3); ($\text{C}_{21}\text{H}_{34}\text{O}$); EIMS (70 eV) m/z $[\text{M}]^+$ 302 (54), 287 (86), 270 (24), 243 (16), 216 (20), 202 (20), 187 (24), 161 (50), 139 (60), 119 (66), 107 (54), 93 (44), 81 (48), 73 (100), 59 (60), 43 (23).

14,18-Epoxyloba-8,10,13(15)-trien-17-ol (11) was obtained as a colorless viscous oil: UV λ_{max} (hexane) 206 (ϵ 4350); $[\alpha]_D +3.8^\circ$ (c 0.38, CHCl_3) ($\text{C}_{20}\text{H}_{32}\text{O}_2$); EIMS (70 eV) m/z $[\text{M}]^+$ 304 (12), 302 (24), 243 (22), 231 (24), 201 (26), 189 (38), 161 (62), 121 (74), 107 (78), 93 (78), 81 (100), 67 (53), 55 (70), 43 (86).

Agar Plate Diffusion Assays. Susceptibility disks (5 mm diameter) were impregnated with 100 μg of the isolated compound and placed on agar plates inoculated with the test bacterium: *B. subtilis* 168 and *S. aureus* ATCC 25923, and *E. coli* ATCC 25922. The plates were checked for inhibition zones after 24 h of incubation at 37 $^\circ\text{C}$.

Bioautographic Detection of Fungicidal Activity. Spores of *C. cucumerinum* were cultivated from carrot-nutrient agar and were inoculated into a liquid yeast culture medium as previously described.⁹ Si gel TLC plates were spotted with the isolated compounds at concentrations of 0.80, 0.40, 0.20, and 0.10 μmol , and the plates were sprayed with a suspension of spores of *C. cucumerinum* in liquid yeast culture medium. The fungitoxic compound was observed as a clear white inhibition spot in a dark layer of the mycelia covering the TLC plate after the inoculated plates were incubated for 2 days at 25 $^\circ\text{C}$.

Brine Shrimp Lethality Test.¹⁰ Eggs of *Artemia salina* (Dohse, Aquaristik GmbH, Bonn, Germany) were hatched in a small tank filled with artificial seawater that was prepared with a commercial salt mixture (Sera Sea-Salt, Aquaristik GmbH, Bonn, Germany) and dis-

tilled water. After 48 h, the phototropic nauplii were collected, 10 shrimps were transferred to each sample vial using a pipet, and artificial seawater was added to make 5 mL. The percent deaths at each dose and control were determined. LC_{50} 's were calculated from the dose-response curve by probit analysis. The experiments were done in triplicate.

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References and Notes

- (1) Shin, J.; Fenical, W. *J. Org. Chem.* **1991**, *56*, 3153–3158.
- (2) Rao, C. B.; Kumar, S. M. D.; Trimurtulu, G.; Rao, D. V. *Indian J. Chem.* **1990**, *29B*, 681–683.
- (3) Hamada, T.; Kusumi, T.; Ishitsuka, M. O.; Kakisawa, H. *Chem. Lett.* **1992**, 33–36.
- (4) Kusumi, T.; Hamada, T.; Ishitsuka, M. O.; Ohtani, I.; Kakisawa, H. *J. Org. Chem.* **1992**, *57*, 1033–1035.
- (5) Dunlop, R. W.; Wells, R. J. *Aust. J. Chem.* **1979**, *32*, 1345–1351.
- (6) Anjaneyulu, V.; Babu, B. H. *Indian J. Chem.* **1993**, *32B*, 1198–1199.
- (7) Raju, B. L.; Subaraju, G. V.; Reddy, K. V.; Rao, C. B. *Indian J. Chem.* **1994**, *33B*, 1033–1037.
- (8) Raju, B. L.; Subaraju, G. V.; Rao, C. B.; Trimurtulu, G. *J. Nat. Prod.* **1993**, *56*, 961–966.
- (9) Gottstein, D.; Gross, D.; Lehmann, H. *Arch. Phytopathol. Pflanzenschutz* **1984**, *20*, 111–116.
- (10) Meyer, B. N.; Ferrigni, N. R.; Putnam, J. E.; Jacobsen, L. B.; Nichols, D. E.; McLaughlin, J. L. *Planta Med.* **1982**, 31–34.

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