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D. Howard Miles, Vallapa Chittawong, Dong-Seok Lho, Allen Matthew Payne, Armando A. de la Cruz, Edgardo D. Gomez, James A. Weeks, and Jerry L. Atwood

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TOXICANTS FROM MANGROVE PLANTS, VII.[1] VALLAPIN AND VALLAPIANIN, NOVEL SESQUITERPENE LACTONES FROM THE MANGROVE PLANT *HERITIERA LITTORALIS*

D. HOWARD MILES,* VALLAPA CHITTAWONG, DONG-SEOK LHO, ALLEN MATTHEW PAYNE,

Department of Chemistry, University of Central Florida, Orlando, Florida 32816

ARMANDO A. DE LA CRUZ,

Department of Biological Sciences, Mississippi State University, Mississippi State, Mississippi 39762

EDGARDO D. GOMEZ,

Marine Science Center, University of the Philippines, Quezon City, Philippines

JAMES A. WEEKS, and JERRY L. ATWOOD

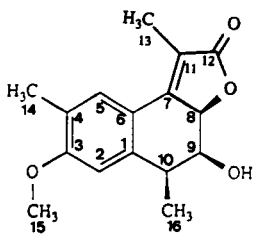
Department of Chemistry, The University of Alabama, University, Alabama 35486

ABSTRACT.—Two novel cadinane sesquiterpenes with an unusual oxygenation pattern and an aromatic ring, vallapin [**1**] and vallapianin [**2**], were isolated from the mangrove plant *Heritiera littoralis*. Vallapin [**1**] showed activity against boll weevils at an inhibition of 80%. The structure was elucidated by spectroscopic methods and X-ray crystallography.

The mangrove plant *Heritiera littoralis* Bryand (Sterculiaceae) has been utilized by natives of the Philippines as a fish poison (2). These observations prompted this continuing chemoecological study of mangrove toxins in the Philippines. An examination of the toxic substances in the petroleum ether and EtOH extracts of *H. littoralis* has been reported (3–5).

In this paper, we report the isolation and structure determination of the compounds vallapin [**1**] and vallapianin [**2**] from *H. littoralis*. Vallapin showed activity against boll weevils at an inhibition level of 80% at a dose of 0.6 mg by the Hedin method (6).

Pure vallapin [**1**] was crystallized from MeOH as white needles [mp 269°; $[\alpha]^{25}D$ −289.5°] from a fraction obtained by chromatography (100% $CHCl_3$) on Si gel. A molecular formula of $C_{16}H_{18}O_4$ was established by hrms ($[M]^+$ *m/z* found 274.1203, calcd 274.1204). This formula indicated eight degrees of unsaturation. The presence of aromaticity in the molecule was suggested by the fact that the molecular ion at *m/z* 274 was also the base peak. Furthermore, fragmentations at *m/z* 245 $[M - CHO]^+$ and *m/z* 246 $[M - CO]^+$ were typical of a phenolic group while fragmentations at *m/z* 77 and *m/z* 128 indicated aromatic



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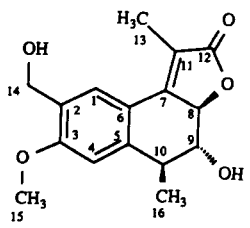

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[1]For Part V, see Miles *et al.* (1). For Part VI, see D.H. Miles, A.M. Ly, V. Chittawong, A.A. de la Cruz, and E.D. Gomez, *J. Nat. Prod.*, **52**, 896 (1989).

and naphthalenic functionalities, respectively.

The presence of an α,β-unsaturated γ-lactone was indicated by the uv (cyclohexane) absorption at 239 nm (ε = 12310, enone group) and by an ir (KBr) band at 1750 cm^{-1}. A band at 3450 cm^{-1} indicated the presence of a hydroxy group. The aromatic nature of vallapin [**1**] was confirmed by the ^1H nmr ($CDCl_3$, 200 MHz), which gave resonances at δ 6.74 (s, 1H, H-4) and 7.48 (s, 1H, H-1) for two isolated protons on an aromatic ring, and by the uv spectrum, which gave absorptions at 217, 286, and 310 nm.

A further study of the ^1H-nmr spectrum showed evidence of the three nonequivalent methyl resonances at δ 1.45 (d, 3H, J = 10 Hz, H_3-13) and 2.31 (s, 3H, H_3-14). Two of the resonances are singlets, proof of their attachment to quaternary carbons. The third methyl group, with double multiplicity, can be assigned to a methine carbon. The ^1H-nmr spectrum also gave signals for a methine at δ 3.06 (m, 1H, H-10), a proton on a carbon bearing oxygen at δ 5.22 (s, 1H, H-8), a proton on C-9 at δ 4.42 (s, 1H, H-9), and a methoxy group at δ 3.95 (s, 3H, H_3-15). Consideration of the spectral data and the isoprene rule led to the assignment of the basic skeleton of vallapin [**1**] as shown. Further justification for this assignment was the fact that structure **1** had the cadinane framework (7,8) which is present in heritol. A single-crystal X-ray structure determination was obtained to verify the structural assignment and to determine stereochemical relationships.

Figure 1 shows a computer-generated perspective drawing of the final X-ray model of vallapin [**1**] less hydrogens. The X-ray experiment defined only the relative configuration, so the enantiomer shown represents an arbitrary choice; however, this enantiomer is probably correct since it has the *R* configuration at C-10 (numbering used in this paper), which is analogous with other cadinanes (7,8).

Vallapianin [**2**] was isolated as a white powder from the 20% MeOH/$CHCl_3$ fraction, mp 182°. A molecular formula of $C_{16}H_{18}O_5$ was determined by hrms ($[M]^+$ *m/z* 290.115). This formula indicated eight degrees of unsaturation. The presence of aromaticity was indicated by the ir bands at 1600 cm^{-1} and 1490 cm^{-1}. The ir bands at 1750 cm^{-1} and 1640 cm^{-1} indicated the presence of an α,β-unsaturated γ-lactone. The aromatic nature of vallapianin [**2**] was confirmed by the ^1H-nmr spectrum, which showed resonances at δ 6.89 (s, 1H, H-

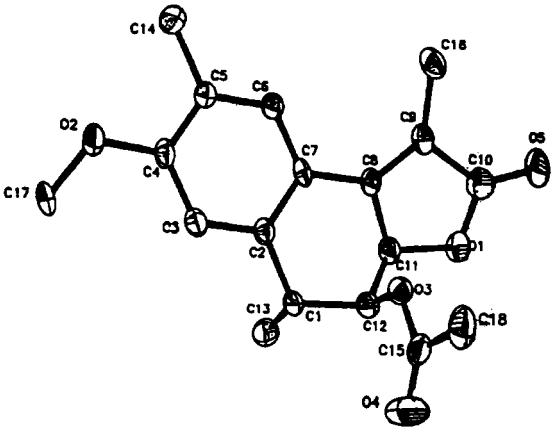

FIGURE 1. Computer-generated perspective drawing of vallapin acetate. Hydrogens are omitted for clarity.

2) and 7.58 (s, 1H, H-5) for two isolated protons on an aromatic ring.

The ^{1}H-nmr spectrum also provided evidence of two non-equivalent methyl resonances at δ 1.55 (d, 3H, J = 7 Hz, H_3-16) and 2.15 (s, 3H, H_3-13). A singlet at δ 2.15 indicated that this methyl group was attached to a quaternary carbon. The doublet at δ 1.55 indicated that this methyl group was attached to a methine carbon. The ^{1}H nmr showed resonances for methylene protons at δ 4.71 (s, 2H, H_2-14), a proton on a carbon-bearing oxygen at δ 4.80 (dd, 1H, J = 1.8 Hz, H-8), a benzylic proton at δ 3.01 (m, 1H, H-10), two hydroxyl groups at δ 3.01 (m, 1H, H-10), two hydroxyl groups at δ 1.23 (14-OH) and 1.60 (9-OH), and methoxy protons at δ 3.91 (s, 3H, H_3-15). The ir spectrum also indicated the presence of hydroxyl groups by an absorption band at 3250–3350 cm^{-1}. These data led to assignment of the basic skeleton of vallapianin [**2**]. Additional justification of this assignment was that the ir spectrum was identical with heritianin [**3**] except for the presence of a much larger band for a hydroxy group. The mass spectrum fragmentation pattern of vallapianin [**2**] also resembled that of heritianin [**3**] (5). The presence of peaks at *m/z* 259, 141, 128, 115, 91, and 77 indicated that the structures of vallapianin [**2**] and heritianin [**3**] were similar. The ^{1}H-nmr spectrum of vallapianin [**2**] was identical with that of heritianin (Table 1) except for the absence of a methyl group at δ 2.25 (s, 3H, H_3-14) and the addition of a methylene group at δ 4.71 (s, 2H, H_2-14). The OH group was placed on C-14 since the methyl group at δ 2.25 in heritianin [**3**] was not present. The structure of vallapianin [**2**] was therefore assigned as the novel structure shown.

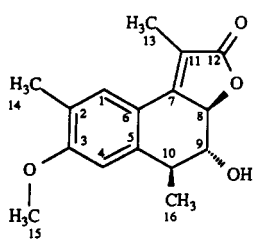

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TABLE 1. Comparison of ^{1}H-nmr Chemical Shifts of Heritianin [**3**] and Vallapianin [**2**].

Proton	Compound	
	3	**2**
H-2	6.85 (s)	6.89 (s)
H-5	7.40 (s)	7.58 (s)
H-8	4.80 (dd, J = 1,8)	4.80 (dd, J = 1,8)
H-9	3.49 (d, J = 7)	3.49 (t, J = 7)
H-10	3.01 (m)	3.01 (m)
H-13	2.15 (s)	2.15 (s)
H-14	2.25 (s)	4.71 (s)
14-OH . . .	—	1.6 (s)
H-15	3.90 (s)	3.91 (s)
H-16	1.55 (d, J = 7)	1.55 (d, J = 7)

EXPERIMENTAL

PLANT MATERIAL AND EXTRACTION.—The roots of *H. littoralis* were collected from the mangrove forest reserve in Pagbilao, Quezon, in Philippines. A voucher specimen, number 80987, has been deposited in the herbarium of the Royal Forest Department, Flora of Thailand. Chopped, air-dried roots of *H. littoralis* (21 kg) were extracted with hexane (21 × 6 liters) in a Soxhlet extractor for 16 h. The hexane extract was removed from the extractor and evaporated in vacuo to yield 98.2 g of crude hexane extract. The defatted roots were allowed to air-dry and were re-extracted with 95% EtOH in a Soxhlet extractor for 16 h. The EtOH extract was concentrated in vacuo to yield 524 g of crude extract. The EtOH extract was partitioned between $CHCl_3$-H_2O (1:1) to yield $CHCl_3$- and H_2O-soluble fractions. The $CHCl_3$ fraction was removed in vacuo to yield 38.4 g of crude extract. The aqueous fraction was freeze-dried to give 70 g of crude extract. The $CHCl_3$-insoluble fraction and the aqueous fraction were filtered to yield 400 g of crude material. The insoluble extract was further partitioned between $CHCl_3$-EtOH (3:1, 1:1, and 1:3) to give four crude extracts, and after concentration in vacuo, the weights of the residues were 40 g, 61 g, 69 g, and 198 g of insoluble material. The $CHCl_3$-EtOH (3:1) extract (20 g) was chromatographed on an open column (diameter 4.5 cm) with 400 g of Si gel as the absorbent. The column was eluted with a hexane/$CHCl_3$/MeOH solvent system.

VALLAPIN [**1**].—The 100% $CHCl_3$ fraction from cc was recrystallized from MeOH to yield 90 mg of vallapin [**1**] as white crystals: mp 269°;

$[\alpha]^{25}D$ −289.5°; ir (KBr) 3450, 1750, 1660, 1620, 1320 cm^{-1}; uv (cyclohexane) λ max 216 (ε 11970), 239 (ε 12310), 286 (ε 16800), 310 (ε 17222); ^1H nmr (200 MHz, $CDCl_3$) δ 1.45 (d, 3H, J = 10 Hz, H_3-16), 2.20 (s, 3H, H_3-13), 2.31 (s, 3H, H_3-14), 3.06 (m, 1H, H-10), 3.95 (s, 3H, H_3-15), 4.42 (s, 1H, H-9), 5.22 (s, 1H, H-8), 6.74 (s, 1H, H-4), 7.48 (s, 1H, H-1); eims (rel. int.) 274 (100), 246 (21), 245 (26), 227 (33), 128 (13), 77 (7); calcd for $C_{16}H_{18}O_4$, 274.3203, found (hreims) 274.3158.

VALLAPIN ACETATE.—Vallapin acetate (mp 180°) was prepared by using vallapin (30 mg), Ac_2O (2 ml), and a few drops of dry pyridine. Crystals of vallapin acetate were grown by slow evaporation of MeOH. Preliminary X-ray photographs showed the orthorhombic symmetry with unit cell parameters a = 7.362 Å, b = 12.290 Å, and c = 18.29 Å; the space group was $P2_12_12_1$ with δ_{calcd} = 1.09 g/cm^3 for Z = 4 (Table 2). Intensities were collected in the usual manner (9) with standard fluctuations of ±2%. The structure was solved by Multan (10) and refined for carbon and oxygen atoms with anisotropic and thermal parameters. Figure 1 shows the molecular structure and numbering scheme.[2]

TABLE 2. Coordinates for Vallapin Acetate.

Atom	x/a	y/b	z/c
C-1	0.284(1)	−0.1026(7)	0.5676(5)
C-2	0.285(1)	0.0081(7)	0.6066(5)
C-3	0.438(1)	0.0390(7)	0.6450(5)
C-4	0.444(1)	0.1386(8)	0.6824(5)
C-5	0.295(1)	0.2072(7)	0.6817(5)
C-6	0.144(1)	0.1768(7)	0.6442(5)
C-7	0.134(1)	0.0782(7)	0.6064(5)
O-1	−0.1935(8)	−0.0726(6)	0.4901(4)
C-8	−0.028(1)	0.0471(7)	0.5633(5)
C-9	−0.163(1)	0.1066(8)	0.5321(5)
C-10	−0.261(1)	0.0300(9)	0.4827(6)
C-11	−0.045(1)	−0.0699(8)	0.5418(6)
C-12	0.137(1)	−0.1113(8)	0.5094(5)
O-2	0.5951(9)	0.1747(5)	0.7193(4)
O-3	0.1752(7)	−0.0411(5)	0.4478(3)
C-13	0.268(2)	−0.1955(7)	0.6239(5)
C-14	0.300(1)	0.3150(7)	0.7231(5)
C-15	0.266(1)	−0.087(1)	0.3907(6)
O-4	0.312(1)	−0.1792(7)	0.3899(5)
O-5	−0.3848(9)	0.0495(7)	0.4425(4)
C-16	−0.211(1)	0.2233(7)	0.5348(6)
C-17	0.727(1)	0.0959(8)	0.7409(6)
C-18	0.295(1)	−0.004(1)	0.3301(5)

[2]Atomic coordinates for this structure have been deposited with the Cambridge Crystallographic Data Centre and can be obtained on request from Dr. Olga Kennard, University Chemical Laboratory, Lensfield Road, Cambridge CB2 1EW, U.K.

VALLAPIANIN [2].—Vallapianin [2] was isolated from the 20% MeOH/$CHCl_3$ fraction from cc. It was recrystallized from Et_2O to yield 60 mg of a white powder: mp 182°; $[\alpha]^{25}D$ 225; ir (KBr) 3250–3350, 1750, 1640, 1600, 1440 cm^{-1}; uv (EtOH) λ max 218 (ε 12111), 239 (ε 13277), 283 (ε 15722), 296 (ε 16444), 302 (ε 16777) nm; ^1H nmr see Table 1; eims *m/z* (rel. int.) $[M]^+$ 290 (48), 272 (66), 259 (13), 243 (28), 213 (100), 141 (11), 128 (17), 115 (16), 91 (8), 77 (7); calcd for $C_{16}H_{18}O_5$, 290.1150, found (hreims) 290.1154.

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LITERATURE CITED

1. D.H. Miles, V. Chittawong, E.D. Gomez, O. de la Cruz-Giron, A.A. de la Cruz, and B.S. Joshi, *J. Nat. Prod.*, **53**, 649 (1989).
2. E. Quisumbing, "Medicinal Plants of the Philippines," Manila Bureau of Printing, 1951, p. 1234.
3. A.A. de la Cruz, E.D. Gomez, D.H. Miles, G.J.B. Cajipa, and V. Chavez, *Int. J. Ecol. Environ. Sci.*, **10**, 1 (1984).
4. D.H. Miles, D.S. Lho, A.A. de la Cruz, E.D. Gomez, J.A. Weeks, and J.L. Atwood, *J. Org. Chem.*, **52**, 2930 (1987).
5. D.H. Miles, A.A. de la Cruz, A.M. Ly, D.S. Lho, E.D. Gomez, J.A. Weeks, and J.L. Atwood. "Toxicants from Mangrove Plants," ACS Symposium Series no. 330, American Chemical Society, Chicago, 1985, pp. 491–495.
6. P.A. Hedin, A.C. Thompson, and J.P. Minyard, *J. Econ. Entomol.*, **59**, 181 (1966).
7. R.M. Coates, *Fortschr. Chem. Org. Naturst.*, **33**, 73 (1976).
8. O. Motl, M. Romanuk, and V. Herout, *Collect. Czech. Chem. Commun.*, **31**, 2085 (1965).
9. J. Holton, M.F. Lappert, D.G. Ballard, R. Pearce, J.L. Atwood, and W.E. Hunter, *J. Chem. Soc., Dalton Trans.*, 45 (1979).
10. G. Germain, P. Main, and M.M. Wolfson, *Acta Crystallogr., Sect. A: Cryst. Phys., Diffr., Theor. Gen. Crystallogr.*, **27**, 368 (1971).

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