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ISOLATION OF PICROPOLYGAMAIN FROM THE RESIN OF *BURSERA SIMARUBA*[1]

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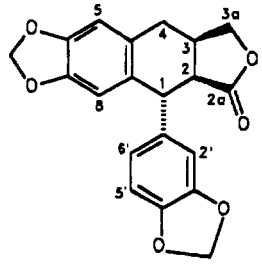
ABSTRACT.—A bioassay-guided fractionation of the resin extract of *Bursera simaruba* resulted in the isolation and identification of the bioactive metabolite picropolygamain [**1**], a 1-aryltetralin lignan having a 2,3-*cis*- lactone ring and two methylenedioxy moieties in its structure.

One of the plants we have investigated as part of our search for bioactive metabolites from Yucatecan medicinal plants is *Bursera simaruba* (L.) Sarg. (Burseraceae), a common tree widely distributed in the tropics. The branches of *B. simaruba* are commonly used by the Yucatecan people as "living" fences. Leaf and/or bark decoctions of this tree are reportedly used in Yucatecan traditional medicine to alleviate the dermatitis caused by the resin of *Metopium brownei* (Jacq.) Urban (Anacardiaceae) (1,2).

Earlier investigations on the chemical constituents of different species from the genus *Bursera* have resulted in the isolation of triterpenes (3–6), bilignans (7), podophyllotoxin-like lignans (8–10), and flavonoids (11). There is, however, only one phytochemical study on metabolites produced by *B. simaruba*, which reports the isolation of elemicine and amyrenol from the resin extract (12).

During our work we found that the $CHCl_3$ extract of the resin of *B. simaruba* showed the presence of biological activity when tested using the brine shrimp assay (13). The extract was subjected to a bioassay-directed fractionation. After successive chromatographic purifications a bioactive fraction showing a single component by tlc and gc was obtained. The biologically active metabolite was obtained as a colorless oil and identified as picropolygamain [**1**], a 1-aryltetranaphthyl lignan lactone previously isolated from the resin of *Commiphora incisa* (Burseraceae) (14).



1

The parent ion peak at *m/z* 352 in the eims of **1** suggested a molecular formula of $C_{20}H_{16}O_6$. This was in agreement with the number of protons and carbons found in the corresponding ^{1}H- and ^{13}C-nmr spectra. The ir spectrum showed aromatic absorption bands at 1615, 1510, and 1490 cm^{-1}, as well as a characteristic lactone-carbonyl band at 1765 cm^{-1}. The presence of a lactone in the structure was confirmed by signals for a carbonyl carbon (178.8 ppm) and a methylene attached to oxygen (73.3 ppm) in the ^{13}C-nmr spectrum. The lack of hydroxyl group absorption bands in the ir spectrum indicated that the remaining four oxygen atoms were present in an ether form. Evidence for the fact that the four oxygen atoms were substituents at aromatic carbons was obtained from the ^{13}C-nmr spectrum where four low-field signals were observed (146.7, 147.2, 147.3, and 148.5 ppm) in addition to three quaternary (128.6, 131.3, and 136.7 ppm) and five

[1]Dedicated to Prof. W.A. Ayer on the occasion of his 60th birthday.

proton-bearing aromatic carbons (108.7, 108.7, 109.4, 110.3, and 121.2 ppm). The ^{13}C nmr also showed that the four oxygens were contained in two methylenedioxy moieties by the presence of two ketal type carbon signals at 101.4 and 101.5 ppm. The lactone, the two methylenedioxy units, and the aromatic rings accounted for twelve of the thirteen unsaturation sites implied by the molecular formula. The remaining unsaturation was assigned to an additional six-membered ring containing a benzylic-type methylene (32.6 ppm) and three methine carbons (33.3, 45.3, and 46.6 ppm). The ^{13}C and ^{1}H chemical shifts were in agreement with those reported for picropolygamain and similar lignan lactones (14–19) and are listed in Table 1.

The ^{1}H-nmr of **1** confirmed the presence of the two methylenedioxy units and allowed for their correct placement in the aromatic rings. Confirmation came from two two-proton overlapping signals, an AB quartet centered at 5.92 (J=1.5 Hz), and a singlet at 5.93. Unequivocal placement of the methylenedioxy moieties at the C-6–C-7 and C-3′–C-4′ positions of **1** was based on a careful analysis of the aromatic region. Three of the protons were part of an ABX system [6.73 (d, J=8.5 Hz, H-5′), 6.62 (dd, J=8.5, 2.7 Hz, H-6′), and 6.60 (d, J=2.7 Hz, H-2′)], and two appeared isolated as a two-proton singlet [6.59 (H-5 and H-8)]. These values were in agreement with those reported for picropolygamain (14) and other lignan lactones having the same substitution pattern in the aromatic rings (10,15–21).

Aryltetralin lignan lactones have been divided according to their having a cis or trans 2,3-lactone ring fusion and to their configuration at C-1 (18,22). It has been reported that cis lactones exist mainly in a boat conformation with the C-1-aryl substituent having a pseudoequatorial

TABLE 1. ^{1}H- and ^{13}C-nmr Data of **1** in $CDCl_3$.

Carbon	$\delta^{13}C$[a]	$\delta^{1}H$
C-1	45.27 (d)	4.36 (d, J=2.9)
C-2	46.60 (d)	3.32 (dd, J=3.1, 9.6)
C-2a	178.83 (s)	—
C-3	33.33 (d)	3.03 (m)
C-3a	73.26 (t)	α=4.44 (dd, J=7.4, 9.2)
		β=3.96 (dd, J=3.3, 9.2)
C-4	32.55 (t)	α=2.82 (dd, J=6.3, 15.3)
		β=2.47 (dd, J=5.1, 15.4)
C-4a	128.64 (s)	—
C-5	108.65 (d)	6.59 (s)
C-6	147.22 (s)	—
C-7	146.74 (s)	—
C-8	109.36 (d)	6.59 (s)
C-8a	131.25 (s)	—
C-1′	136.66 (s)	—
C-2′	108.72 (d)	6.60 (d, J=2.7)
C-3′	148.49 (s)	—
C-4′	147.30 (s)	—
C-5′	110.32 (d)	6.73 (d, J=8.5)
C-6′	121.19 (d)	6.62 (dd, J=2.7, 8.5)
OCH_2O-	101.44 (t)	5.93[b] (s)
OCH_2O-	101.57 (t)	5.92[b] (ABq, J=1.5)

[a]Multiplicities determined from APT experiment.

[b]Chemical shift values may be exchanged.

orientation and the H-1 proton occupying the flagpole position (18). The chemical shift (4.36 ppm) and $J_{1,2}$ (2.9 Hz) values observed for H-1 in the ^{1}H nmr of **1** were in agreement with those reported for a cis lactone having a 1,2-*trans*-2,3-*cis* stereochemical arrangement (17–19). On the basis of the data presented and by comparing it with that reported in the literature it was possible to identify **1** as picropolygamain.

It is well known that lignans display a wide range of biological activities including antitumor, antimitotic, and antiviral (23,24). When tested in the brine shrimp assay, the LC_{50} of **1** was found to be 52.2 ppm. Further in vitro evaluation against three human tumor cell lines indicated that **1** has cytotoxic activity comparable to that of adriamycin (Table 2) and is worthy of further evaluation.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—The ir spectrum was recorded in $CHCl_3$ using a Perkin-Elmer 683 spectrophotometer. The low resolution ms was measured using a Hewlett-Packard 5890 gas chromotograph (column Ultra 2, Crosslinked 5% Ph Me Silicone, 25 m×0.32 mm×0.52 μm film thickness; oven temperature 265°) coupled to a Hewlett-Packard 5971A mass selective detector. ^{1}H (200 MHz) and ^{13}C nmr (50 MHz) were recorded in a Varian GEM-200 spectrometer, using $CDCl_3$ as the solvent and TMS as internal standard. Tlc was carried out using precoated Si gel aluminum plates (E. Merck DC Alufolien, Kieselgel 60 F_{254}, 0.2 mm thickness). Chromatograms were examined under uv light using a uv-viewing cabinet (Spectroline Model CX-20) and by spraying with 4% phosphomolybdic acid containing a trace of ceric sulfate in 5% H_2SO_4. Flash cc was run using Si gel 60 (230–400 mesh) from Aldrich Chemical Co.

PLANT MATERIAL.—The resin was collected in January 1991 from trees growing on the grounds of CICY in Merida, Yucatan, Mexico. A voucher specimen (LP-2) has been deposited in the herbarium of CICY. The exudate was obtained by making cuts in the bark and soaking the resin in a previously weighted piece of cotton.

EXTRACTION AND ISOLATION.—Soxhlet extraction of the exudate (35 g) using $CHCl_3$ (400 ml) resulted in 16.9 g (48.2%) of crude resin extract. The extract was chromatographed [flash cc, hexane-Me_2CO (95:5)] yielding 22 mixed fractions. Further purification [flash cc, C_6H_6-Me_2CO-MeOH (94:4:2)] of the active fraction (363.5 mg) gave 12 new fractions, two of them showing activity in the brine shrimp assay. Final purification of fractions BSR-44B [15.8 mg, flash cc, hexane-Me_2CO (7:3)] and BSR-44C [42.6 mg, flash cc, C_6H_6-EtOAc (9:1)] yielded 18.8 mg (0.11%) of pure **1** as a colorless oil: R_f 0.37 [hexane-EtOAc (7:3)]; ir ($CHCl_3$) 3030 (s), 2780 (w), 1765 (s), 1615 (w), 1510 (s), 1490 (s), 1235 (s), 1040 (s), 945 (m), 930 (m), 870 (w), 850 (w) cm^{-1}; eims m/z (rel. int.) [M]$^+$ 352 (100), 307 (7.7), 292 (11.6), 280 (32.2), 268 (22.7); ^{1}H and ^{13}C nmr see Table 1.

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LITERATURE CITED

1. J.F. Morton, "Atlas of Medicinal Plants of Middle America," Charles C Thomas, Illinois, 1981.
2. E. Cabrera, M. Sousa, and O. Tellez, "Imagenes de la flora quintanarroense," CIQRO, Quintana Roo, Mexico, 1982.
3. K.J. Crowley, *J. Chem Soc.*, 4254 (1964).
4. I.C. Parsons, A.I. Gray, C. Lavaud, G. Massiot, and P.G. Waterman, *Phytochemistry*, **30**, 1221 (1991).
5. K.V. Syamasundar, G.R. Mallavarapu, and E.M. Krishna, *Phytochemistry*, **30**, 362 (1991).

TABLE 2. Cytotoxic Activities of **1** and Adriamycin.

Compound	Cell Culture (ED_{50} μg/ml)		
	A-549 (lung)	MCF-7 (breast)	HT-29 (colon)
1	$<10^{-2}$	$<10^{-2}$	$<10^{-2}$
Adriamycin	4.09×10^{-4}	1.05×10^{-4}	1.12×10^{-4}

6. S.D. Jolad, R.M. Wiedhopf, and J.R. Cole, *J. Pharm. Sci.*, **66**, 889 (1977).
7. J.D. Hernandez, L.U. Roman, J. Espiñeira, and P. Joseph-Nathan, *Planta Med.*, **47**, 215 (1983).
8. E. Bianchi, M.E. Caldwell, and J.R. Cole, *J. Pharm. Sci.*, **57**, 696 (1968).
9. P.B. McDoniel and J.R. Cole, *J. Pharm Sci.*, **61**, 1992 (1972).
10. S.D. Jolad, R.M. Wiedhopf, and J.R. Cole, *J. Pharm. Sci.*, **66**, 892 (1977).
11. M.P. Souza, M.I.L. Machado, and R. Braz-Filho, *Phytochemistry*, **28**, 2467 (1989).
12. R. Pernet, *Lloydia*, **35**, 280 (1972).
13. B.N. Meyer, N.R. Ferrigni, J.E. Putnam, L.B. Jacobsen, D.E. Nichols, and J.L. McLaughlin, *Planta Med.*, **45**, 31 (1982).
14. G.J. Provan and P.G. Waterman, *Planta Med.*, 271 (1985).
15. G.M. Sheriha, K. Abouamer, B.Z. Elshtaiwi, A.S. Ashour, F.A. Abed, and H.H. Alhallaq, *Phytochemistry*, **26**, 3339 (1987).
16. J. Jakupovic, A. Schuster, F. Bohlmann, R.M. King, and H. Robinson, *Planta Med.*, 18 (1986).
17. A. San Feliciano, M. Medarde, J.L. Lopez, P. Puebla, J.M. Miguel del Corral, and A.F. Barrero, *Phytochemistry*, **28**, 2863 (1989).
18. S.P. Forsey, D. Rajapaksa, N.J. Taylor, and R. Rodrigo, *J. Org. Chem.*, **54**, 4280 (1989).
19. A. San Feliciano, J.M. Miguel del Corral, M. Gordaliza, and A. Castro, *Phytochemistry*, **29**, 1335 (1990).
20. Y. Al-Abed, S. Sabri, M.A. Zarga, Z. Shah, and Atta-ur-Rahman, *J. Nat. Prod.*, **53**, 1152 (1990).
21. R. Stevenson and J.V. Weber, *J. Nat. Prod.*, **52**, 367 (1989).
22. P.M. Dewick and D.E. Jackson, *Phytochemistry*, **20**, 2277 (1981).
23. W.D. MacRae and G.H.N. Towers, *Phytochemistry*, **23**, 1207 (1984).
24. W.D. MacRae J.B. Hudson, and G.H.N. Towers, *Planta Med.*, **55**, 531 (1989).

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