

Research Note

SOME CONSTITUENTS OF MABI BARK¹

Both *Colubrina arborescens* (Mill.) Sarg and *C. elliptica* (Sw.) Brinz and Stern (Rhamnaceae) are known as mabi. The extracts of the tree bark are used as folk remedies and in popular beverages throughout the Caribbean region². Saponins have been found in *C. arborescens*³. A range of chemical compounds have been estimated in the fermented drink derived from *C. elliptica*^{4,5}. A variety of chemical substances have been found in other species of the genus *Colubrina*^{6,7,8,9,10}. This paper presents the results of the identification of the tannins, alkaloids and saponins in mabi bark, since these con-

stituents are likely to be the sensory or physiologically-active agents in mabi drinks.

The methanol soluble extractives of the mabi bark were fractionated by being shaken sequentially with petroleum spirit, dichloromethane, ethyl acetate and n-butanol. The largest fraction (9% of the total) was found in the butanolic extract, and it contained a mixture of saponins, which were separated by repeated preparative thin layer chromatography on silica gel, with various chloroform-methanol solvent systems. These saponins are bitter tasting

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²Morton, J. F., 1981. Atlas of medicinal plants of Middle America. C. C. Thomas, Illinois, U.S.A.

³Colard, M. J. M., P. A. Dumont and F. Campernolle, 1975. Sugar sequence analysis by mass spectrometry of a new saponoside isolated from the bark of *Colubrina arborescens* Mill., *Biomed. Mass Spectrom.* 2:156-63.

⁴Montoya, A. and H. D. Graham, 1981. Microbiological studies of mabi fermentation. *J. Agric. Univ. P. R.* 65:219-31.

⁵Graham, H. D. and P. E.L. Zengotita, 1982. Composition of mabi ingredients, mabi prepared by the conventional method and by controlled fermentation, *J. Agric. Univ. P. R.* 66:293-99.

⁶Wani, M. C., H. L. Taylor and M. E. Wall, 1973. Plant antitumour agents. XII. Texensine, a New Peptide Alkaloid from *Colubrina texensis*, *Tetrahedron Lett.* (47):675-678.

⁷Wani, M. C., H. L. Taylor and M. E. Wall, 1973. Plant antitumour agents: colubrinol acetate and colubrinol, antileukaemia ansa macrolides from *Colubrina texensis*, *J. Chem. Soc. Chem. Comm.* 390.

⁸Roitman, J. N. and L. Jurd, 1978. Triterpenoid and phenolic constituents of *Colubrina asiatica*, *Phytochem.* 17:491-94.

⁹Guinaudeau, H., O. Seligman, H. S. Wagner and A. Neszmelyi, 1981. Faralaoiroside and faratroside, two flavonoid triglycosides from *Colubrina faralaostra*, *Phytochem.* 20:1113-116 (and literature cited therein).

¹⁰Wagner, H. S., S. Otto, K. Jurcic, J. F. Morton and A. Neszmelyi, 1983. Chemistry, carbon-13 NMR study and pharmacology of two saponins from *Colubrina asiatica*, *Planta Med.* 48:136-41.

compounds and are notoriously difficult to purify¹¹. The major saponin sub-fraction was subjected to acidic hydrolysis and yielded sugars and mainly the triterpenoid saponin ebelin lactone, $C_{30}H_{46}O_3$. This was established by UV, IR, NMR and mass spectrometry,¹² and comparison with findings in the literature^{10,11}. It is likely that ebelin lactone was produced during the hydrolysis reaction from the acid-labile triterpenoid jujubogenin $C_{30}H_{48}O_4$, and that jujubogenin may be the genuine aglycone in the saponins from mabi bark. Earlier workers had found jujubogenin glycosides as the saponins in the leaves of *Colubrina asiatica*^{10,11}.

The total phenolic content of mabi bark was estimated by the Folin-Denis method⁶ as 4%. The tannin content based on aqueous-acetone extracts of the bark was estimated as 0.15% by the method of Seigler et al.¹³ The tannins are astringent because they precipitated proteins such as gelatin. Boiling the aqueous extract of mabi bark with 2M HCl for 30 minutes yielded the anthocyanidin delphinidin: therefore the tannins appear to be prodelphinidin and cyanidin compounds¹⁴. No gallic acid derivatives (gallotannins) were found.

The ground mabi bark was alkalinized with 20% sodium carbonate aqueous solution and then percolated with chloroform. The chloroform-solubles (0.001% of the bark) were purified by solvent extractions and by preparative thin layer chromatography with ethyl acetate-chloroform (2:1) on silica gel, to yield a mixture of alkaloids. Two components (at R_f 0.37 and 0.20) were separated and re-crystallized from ethyl acetate. Their spectral data were obtained, and each was hydrolyzed exhaustively with 6 M HCl at 100° for 24 hours in a sealed tube. They were shown to be the cyclopeptides known as scutianine B¹⁵ and acutianine C¹⁶, respectively. These alkaloids have been found in other plants of the Rhamnaceae family.

On the basis of the relative solubilities in water, the alkaloids are expected to contribute much less than the bitter-tasting saponins and the astringent tannins to the special taste of mabi drinks.

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¹¹Hostettman, K., M. Hostettman and A. Marston, 1984. Isolation of natural products by droplet counter current chromatography and related methods, *Nat. Prod. Reports*, 1:471-81.

¹²Mass spectral data were obtained by the courtesy of the analytical services of the International Organization for Chemical Sciences in Development of UNESCO.

¹³Seigler, D. S., S. Seilheimer, J. Keesy, H. F. Huang, 1986. Tannins from four common *Acacia* species of Texas and North Eastern Mexico, *Econ. Bot.* 40:220-32.

¹⁴Harborne, J. B., 1984. *Phytochemical Methods*. 2nd ed, Chapman and Hall, U. K.

¹⁵Tschesche, R., E. Ammermann, and H. Fehlhäber, 1971. Alkaloide aus Rhamnaceen X. Scutianine-B ein weiteres Peptidalkaloid aus *Scutia buxifolia* Reiss, *Tetrahed. Lett.* (46):4405-4408.

¹⁶Tschesche, R. and Ammermann, E., 1974. Alkaloide aus Rhamnaceen XXIII. Scutianine-C, -D und -E, drei weitere Cyclopeptidalkaloide aus *Scutia buxifolia* Reiss. *Chem. Ber.* 107:2274-283.