

5,6-DIMETHOXY-7-HYDROXYCOUMARIN (UMCKALIN) FROM *ALLAMANDA BLANCHETTI*: ISOLATION AND ^{13}C -NMR CHARACTERISTICS

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The EtOH extract of the root bark of *Allamanda blanchetti* A. DC. (Apocynaceae) was fractionated according to the method used for *Allamanda cathartica* by Kupchan *et al.* (1). The CCl_4 extract, after usual work up followed by column chromatography, tlc, and crystallization, yielded, in addition to the iridoid lactones, plumericin and isoplumericin, a very light yellow coumarin, $\text{C}_{11}\text{H}_{10}\text{O}_5$ (M^+ 222), mp 149-150°. This isolate proved to be 5,6-dimethoxy-7-hydroxycoumarin, or umckalin, (2,3) by spectral (uv, ir, ^1H and ^{13}C nmr, ms) and chemical evidence. The physical properties (mp, uv, ir, ^1H nmr, ms) of the coumarin and its methyl derivative are very close to those of umckalin and its methyl derivative, respectively. Also, the ^{13}C -nmr spectra of the coumarin and its methyl ether (Table 1), not reported earlier, are consistent with the umckalin structure. This is the first report of the isolation of umckalin from any other source outside of the genus *Pelargonium*.

EXPERIMENTAL

MATERIALS.—The plant material was collected from an area within 50 km of Joao Pessoa, and the herbarium specimen is kept in the LPX Herbarium of the Universidade Federal da Paraiba. The mps were taken in a Kofler hot stage and are uncorrected. The uv spectra were determined in a Carl Zeiss Jena VSU-2-P spectrophotometer, and the ir spectra were obtained in a Perkin-Elmer 467 grating spectrophotometer. Ms was determined in a Hewlett-Packard 5930 quadropole mass spectrometer, and the nmr spectra were recorded in a Varian FT 80A instrument. Silica gel (E. Merck, No. 7734) was used for analytical tlc plates and silica gel (E. Merck, 60 PF₂₅₄) was used for preparative tlc.

EXTRACTION AND PURIFICATION OF THE CONSTITUENTS.—The 95% EtOH extract (108.7 g) of the roots of *A. blanchetti* (600 g) was fractionated according to the method of Kupchan *et al.* (1). The CCl_4 extract was chromatographed over a column of silica gel, and C_6H_6 was used as eluent yielding plumericin and a mixture of isoplumericin and umckalin. The latter mixture was separated by preparative tlc. Plumericin and isoplumericin were identified by comparison with their physical properties (mp, uv, ir, ms, and ^1H nmr) with those published in the literature (1). Identification of umckalin was made by comparison of the physical characteristics (mp, uv, ir, ms, and ^1H nmr) of the compound and its methyl ether with those published in the literature (2,3) and was confirmed by ^{13}C -nmr spectroscopy (Table 1). No sample was available for direct comparison. Details are available upon request to the senior author.

TABLE 1. ^{13}C -nmr Chemical Shifts^a (δ) of Umckalin and Its Methyl Ether

Carbon Atom	Umckalin (5,6-dimethoxy-7-hydroxycoumarin)	Umckalin Methyl Ether (5,6,7-Trimethoxy-coumarin)
2	161.7	161.2
3	112.2	112.6
4	138.9	138.9
5	151.6	151.7
6	136.7	138.3
7	153.9	157.3
8	99.0	95.7
9	148.7	149.4
10	107.2	107.3
OCH ₃	61.2, 61.5	56.3, 61.2, 61.8

^aIn CDCl_3/TMS .

ACKNOWLEDGMENTS

We thank the NPPN, Universidade Federal do Rio de Janeiro, for some of the ms. Thanks are due to

Sra. Maria de Fatima Agra for the collection and Prof. Valerio F. Ferreira, Botanical Garden, Rio de Janeiro, for the identification of the plant material.

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Received 8 November 1984

CHEMICAL CONSTITUENTS OF THE ROOTS OF *ACANTHUS ILLICIFOLIUS*

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In our search for biologically active substances from mangrove plants that grow in Thailand, we report here the isolation of octacosyl alcohol, stigmasterol, benzoxazoline-2-one, and stigmasteryl- β -D-glucopyranoside from the roots of *Acanthus illicifolius* L. (Acanthaceae). The roots have been shown (1) to have activity against Friend leukemia virus in erythroleukemic Swiss mice. Benzoxazoline-2-one has been extensively investigated (2) for medicinal value as a central nervous system depressant, exhibiting analgesic, antipyretic, anticonvulsant, hypnotic, and muscle relaxant activity. Benzoxazoline-2-one is also reported (2) to be a resistance factor for fungi. Furthermore, there are reports (3) that ribose derivatives of this compound are active as anticancer and antiviral agents. Stigmasterol has been shown (4) to have a slight hypercholesterolemic effect while exhibiting no obvious effect on the heart or liver.

There have been a number of phytochemical investigations (5-7) with *A. illicifolius*; however, this is the first report of the presence of octacosyl alcohol, benzoxazoline-2-one, and stigmasteryl- β -D-glucopyranoside in this species.

EXPERIMENTAL

PLANT MATERIAL.—The plant used in this study was identified as *A. illicifolius* by Dr. Pipat Patanongpaiboon, Department of Botany, Chulalongkorn University, and representative specimens are being maintained in the Department of Botany at Chulalongkorn University.

EXTRACTION.—The roots of *A. illicifolius* were collected in Samuthsakorn, Thailand, in April 1982, (6 kg), dried, and extracted with petroleum ether. The solvent was removed in vacuo and subsequently chromatographed on silica gel using the quick column technique (8) to obtain octacosyl alcohol (253 mg) and stigmasterol (400 mg).

The plant material remaining after extraction with petroleum ether was extracted with 70% EtOH at room temperature. Using standard solvent, solvent and chromatographic procedures (9-11), benzoxazoline-2-one (472 mg) and stigmasteryl- β -D-glucopyranoside (400 mg) were obtained.

All compounds were identified by standard spectral means, by the formation of derivatives, and by ir, uv, and nmr ^1H nmr comparison with an authentic sample.

Full details of the isolation and identification of the compounds are available on request to the senior author.

ACKNOWLEDGMENTS

The assistance of Dr. P. A. Hedin (USDA Boll Weevil Research Laboratory) in obtaining the mass spectra is appreciated.

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