

The metabolites produced by several species of *Nectria* have been reported previously (4). 2,5-Dimethoxy-3,6-dimethylbenzoquinone is a known metabolite of *Nectaria coryli* (2), whereas this is the first reported isolation of (-)-mellein from a species of this genus.

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COUMARINS FROM BARK OF *AMYRIS BARBATA*

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Species of *Amyris* (Rutaceae) are known to be rich sources of coumarins, and recently some quinoline alkaloids and novel nicotinamide derivatives have been reported (1,2). *Amyris barbata* Lundell is a tree that grows in the premontane forest of Costa Rica, constituting a precious wood for its durability and resistance. It has been used in traditional medicine for the treatment of skin diseases (3)¹. We have examined the bark of *A. barbata* and report, in addition to β -sitosterol and *p*-anisic acid, three known coumarins: scopoletin, imperatorin, and oxyimperatorin. The compounds were characterized by comparison with literature data (4-6) or with authentic samples.

EXPERIMENTAL

PLANT MATERIAL.—The material was collected in December in Santa Ana, Province of San Jose, Costa Rica, and identified by L.J. Poveda; voucher specimens have been deposited at the Herbarium of the National Museum, No. 108523. Uv spectra were recorded in MeOH; ¹H-nmr and ¹³C-nmr spectra were recorded with TMS as internal standard in CDCl₃. Si gel 60 (Merck, 0.040-0.060 mm) was used for column chromatography and Si gel G (Merck) for tlc.

ISOLATION OF CHEMICAL CONSTITUENTS.—Bark of *A. barbata* (1.47 kg) was extracted with MeOH at room temperature and concentrated under reduced pressure to yield a crude extract (52 g), which was partitioned with hexane (20.6 g) and EtOAc (5.17 g). The concentrated hexane extract (10 g) was fractionated by flash chromatography (500 g Si gel, 50 ml fractions); 100 fractions eluted with hexane-Et₂O (1:1) were collected, and each was analyzed by tlc. Fractions 17-32, after successive recrystallizations from MeOH, afforded β -sitosterol (0.50 g) identified by standard sample comparison (mmp, ir, ¹H nmr, tlc). Fractions 89-100 were combined and subjected to preparative tlc on Si gel using hexane-EtOAc (3:1), which yielded imperatorin (0.02 g). The EtOAc concentrate (5.17 g) was fractionated by flash chromatography (300 g Si gel, 25 ml fractions) into 82 fractions using hexane-EtOAc (3:1) as eluent. Fractions 11-48 were rechromatographed over Si gel with hexane-EtOAc (3:1) which yielded scopoletin (0.33 g, ¹H nmr, ¹³C nmr, mp) (7) and imperatorin (0.70 g, uv, ¹H nmr, mp) (8, 9). Fractions 49-82 were purified by using the same chromatography system as above but with CHCl₃-MeOH (95:5) as eluent to give imperatorin oxide (0.61 g, ¹H nmr, ¹³C nmr, mp) (10), scopoletin (0.05 g), and *p*-anisic acid (0.015 g), which was identified by mp, co-tlc and superposable ir with an authentic sample obtained by treating *p*-anisic-acid methyl ester with base (NaOH 1N) at room temperature for 1 h. Details of the identification are available from the major author.

¹L.J. Poveda, personal communication, 1985.

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