



DIHYDROCHALCONES, COUMARINS AND ALKALOIDS FROM *METRODORA NIGRA**

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Key Word Index—*Metrodorea nigra*; Rutaceae; prenylated dihydrochalcones; steroids; lignan; furoquinoline alkaloids; coumarins.

Abstract—Two novel dihydrochalcones, 2',3,4',6'-tetrahydroxy-4-methoxy-3',5-di-(3,3-dimethylallyl)-dihydrochalcone and 2',3,6'-trihydroxy-4-methoxy-5-(3,3-dimethylallyl)-3',4'-(2'',2''-dimethyldihydropyran)-dihydrochalcone, have been isolated from fresh fruits of *Metrodorea nigra*. Stems and leaves showed a similar composition and we have isolated common steroids, simple coumarins, several furocoumarins, furoquinoline alkaloids and a furofuran lignan. From stems, we have also isolated the pentacyclic 6-C-monoterpenyl-5,7-dioxycoumarin, deoxybruceol. Structures of the isolated compounds were elucidated on the basis of spectral data.

INTRODUCTION

The genus *Metrodorea*, belonging to the tribe Cusparieae, is considered to contain *ca* eight species, which are confined to Brazil [1, 2]. There are, as yet, no reports of any chemical studies on *Metrodorea* species. Thus, as part of our ongoing phytochemical and chemotaxonomic study on members of the Cusparieae, we have examined fresh fruits, leaves and stems of *M. nigra*. This species, commonly known as wild orange, is a small tree of 1–2 m high. We report here in the isolation and characterization of two new dihydrochalcones, along with known coumarins, anthranilate alkaloids, lignans and steroids.

RESULTS AND DISCUSSION

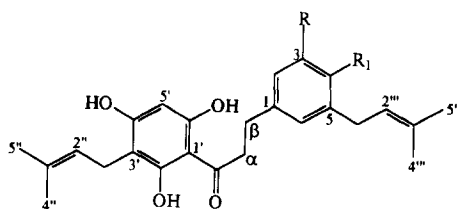
The fresh fruits of *M. nigra* were percolated with methanol. The chloroform-soluble fraction of the methanol extract was concentrated and subjected to VLC. Apolar and polar fractions were composed mainly by triacyl glycerols and condensed tannins, respectively. From the medium polar fractions, after prep. TLC, we obtained two novel C-prenylated dihydrochalcones. Compound 1 exhibited similar spectral data to 2',4,4',6'-tetrahydroxy-3,5-di-(3,3-dimethylallyl)-dihydrochalcone (6), which has previously been isolated from *Boronia inconspicua* (Rutaceae) [3]. The ¹H NMR spectrum (Table 1), instead of signals for *meta*-coupled A-ring protons,

showed one 1H singlet at δ 5.80. This signal and the mass spectral fragment at *m/z* 220 [C₁₂H₁₂O₄]⁺, due to fission of the propanone bridge between the C = O and the C-α, established the presence of a 3,3-dimethylallyl group at C-3'/C-5'. The signal at δ 5.80 was thus assigned to H-5'/H-3'. This was supported by comparison with the spectral data published for 5,7,2',4'-tetrahydroxy-6,5'-di-(3,3-dimethylallyl)-flavanone, isolated from *Euchresta japonica* (Leguminosae) [4]. The ¹H NMR also showed a B-ring spin-system for 3,4,5-substitution, as in 6, and a signal for a methoxyl group. However, for 1, H-6 (δ 6.67) was more deshielded than that at H-2 (δ 6.54) and, in the ¹³C NMR spectrum, the deshielded position of the methoxyl resonance at δ_C 61.2 (*q*) required an *ortho*-disubstituted carbon [5], thus placing the methoxyl substituent at C-4, the hydroxyl substituent at C-3 and the 3,3-dimethylallyl substituent at C-5. This was corroborated by the mass fragment *m/z* 218 [C₁₄H₁₈O₂]⁺. Thus, 1 was identified as 2',3,4',6'-tetrahydroxy-4-methoxy-3',5-di-(3,3-dimethylallyl)-dihydrochalcone.

Compound 2 was isolated in small amounts. The ¹H NMR spectrum (Table 1) was similar to that of 1 for the A and B-ring protons, consequently, the same substitution pattern was deduced for 2. However, the ¹H NMR spectrum also indicated the presence of one 2,2-dimethyldihydropyran ring (δ 1.36, 6H, *s*; 1.80, 2H, *t*, *J* = 6.6 Hz and 2.58, 2H, *t*, *J* = 6.6 Hz). These observations can be explained only by the presence of a 2,2-dimethyldihydropyran ring between C-3' and O-4'. This was confirmed by the mass fragment at *m/z* 220 (C₁₂H₁₂O₄)⁺ as found in 1. Thus, 2 was identified as 2',3,6'-trihydroxy-4-methoxy-5-(3,3-dimethylallyl)-3',4'-(2'',2''-dimethyldihydropyran)-dihydrochalcone. However, compound 2 could be an artefact formed from 1 during isolation.

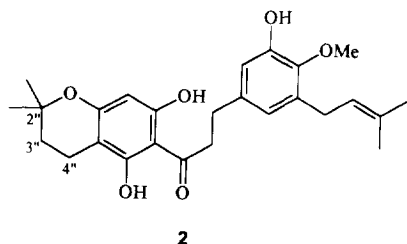
*Based in part on the doctorate thesis presented by the senior author to the Universidade Federal de São Carlos, SP, Brazil (1994).

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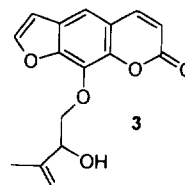


1 R = OH, R₁ = OMe

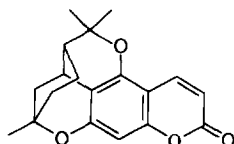
6 R = 3,3-dimethylallyl, R₁ = OH



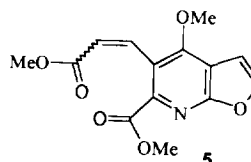
2



3



4



5

Stems and leaves of *M. nigra* were percolated with hexane, chloroform and methanol, respectively. The hexane and chloroform extracts, and the chloroform-soluble fractions of the methanol extracts, were investigated. From the leaves, we isolated the coumarins, limetin [6], aurapten [7], 8-methoxylimetin [8], bergapten [9], xantotoxin [10], isopimpinellin and isoimperatorin [11], the furoquinoline alkaloid, skimmianine [12], the furofuran lignan, eudesmin [13] and a mixture of β -sitosterol and stigmasterol. From the stems we isolated the coumarins, limetin, aurapten, 8-methoxylimetin, bergapten, xantotoxin, isopimpinellin, imperatorin [11], scopoletin [14], umbelliferone [14], isogospherol (3) [15] and deoxybruceol (4) [16–18], the furoquinoline alkaloid, skimmianine, the secofuroquinoline alkaloids, *E*- and *Z*-dimethylrhoifolinol (5) [19], and a mixture of β -sitosterol and stigmasterol. These compounds were identified by comparison of their spectral data with those reported in the literature. Isogospherol (3), a 8-modified *O*-prenylated linear furocoumarin previously isolated from *Prangos lophoptera* (Umbelliferae) [15], does not appear to have previously been recorded from the Rutaceae. Also, this appears to be the first record of deoxybruceol (4), a pentacyclic 6-*C*-monoterpenyl-5,7-dioxycoumarin, from the tribe Cusparieae, but it has been reported from *Eriostemon brucei* (tribe Boronieae) [16, 17]. The furoquinoline alkaloids, *Z*- and *E*-dimethylrhoifolinol (5), have been reported previously by us from another Braz-

ilian species of the Rutaceae, *Zanthoxylum rhoifolium* [19]. The range of compounds we have isolated from *M. nigra* reinforces its position as a member of the Rutaceae [20].

EXPERIMENTAL

General. Mps uncorr. UV and IR were recorded in MeOH and CHCl₃, respectively. ¹H and ¹³C NMR spectra were recorded at 200 and 50 MHz, respectively, in CDCl₃ with TMS as int. standard. EIMS were obtained at 70 eV (probe).

Plant material. *Metrodorea nigra* St. Hil. was collected in the State of São Paulo, Brazil, in April 1984. A voucher specimen is deposited at the Herbarium of the Instituto Agrônomo de Campinas, SP, Brazil.

Extraction and isolation. Fresh fruits (110 g) were chopped into small pieces and percolated with MeOH. The residue was dried and extracted with CHCl₃ at room temp. This extract (2.5 g) was shown to be composed of satd hydrocarbons and triacylglycerols. The MeOH extract (10 g) was suspended in 50% aq. MeOH and partitioned between CHCl₃, EtOAc and *n*-BuOH. The CHCl₃-sol. fr. (2.2 g) was submitted to VLC over silica gel 60 H using CH₂Cl₂–MeOH mixts of increasing polarity and, after combination, monitored by TLC, furnished 5 frs (A–E). Fr. B (42 mg), prep. TLC (silica gel;

Table 1. NMR data for compounds **1** and **2**

H/C	1		2
	H	C	H
1	—	138.5 s	—
2	6.54 d (2.0 Hz)	113.2 d	6.58 d (2.2 Hz)
3	—	143.2 s	—
4	—	148.6 s	—
5	—	113.0 s	—
6	6.67 d (2.0 Hz)	122.7 d	6.70 d (2.2 Hz)
1'	—	104.9 s	—
2'	—	160.1 s*	—
3'	—	105.6 s	—
4'	—	162.5 s	—
5'	5.80 s	95.3 d	5.93 s
6'	—	160.8 s*	—
C=O	—	204.8 s	—
α	2.86 t (7.2 Hz)	30.4 t	2.89 t (7.2 Hz)
β	3.31 t (7.2 Hz)	45.6 t	3.34 t (7.2 Hz)
1''	3.31 m	21.6 t	—
2''	5.23 m	121.6 d**	—
3''	—	132.6 s***	1.80 t (6.6 Hz)
4''	1.72–1.81	17.9 q	2.58 t (6.6 Hz)
5''	1.72–1.81	25.8 q	—
1'''	3.31 m	28.2 t	3.34 m
2'''	5.23 m	121.7 d**	5.23 m
3'''	—	134.4 s***	—
4'''	1.72–1.81	17.9 q	1.73 s
5'''	1.72–1.81	25.8 q	1.73 s
4-OMe	3.76 s	61.2 q	3.77 s
2 × (2''-Me)	—	—	1.36 s
OH	5.64 br s	—	5.45 br s
OH	6.21 br s	—	5.45 br s
OH	8.70 sh	—	—
OH	13.80 s	—	13.77 s

The number of asterisks indicates interchangeable signals.

Values in parentheses are coupling constants. Multiplicities of carbons were determined using DEPT.

hexane-CH₂Cl₂-EtOAc-MeOH, 15:7:7:1, 2 runs), afforded triacylglycerols (15 mg), **1** (19 mg) and **2** (1 mg). The other frs showed a complex composition. EtOAc (0.2 g) and *n*-BuOH (0.8 g) frs were composed mainly of sugars.

Stems (284 g) were ground and percolated with hexane, CHCl₃ and MeOH, respectively. Similar chromatographic treatment as described for fruits gave, in order, aurapten (21 mg), limetin (13 mg), a mixt. of β -sitosterol and stigmaterol (22 mg), bergapten (3 mg), xantotoxin (11 mg), imperatorin (4 mg), deoxybruceol (**4**) (0.9 mg), isopimpinellin (10 mg), 8-methoxylimetin (6 mg), skimmianine (1.5 mg), *E,Z*-dimethylrhoifolinate (**5**) (0.8 mg), isogospherol (**3**) (0.5 mg), scopoletin (8 mg), umbelliferone (6 mg) and 8-methoxylimetin (53 mg).

Leaves (850 g) were ground and percolated with hexane, CHCl₃ and MeOH, respectively. Chromatographic treatments similar to those used for fruits gave, in order, aurapten (19 mg), a mixt. of β -sitosterol

and stigmaterol (23 mg), limetin (12 mg), bergapten (4 mg), xantotoxin (3 mg), isopimpinellin (5 mg), 8-methoxylimetin (6 mg), isoimperatorin (5 mg), skimmianine (10 mg) and eudesmin (17 mg).

2',3,4',6'-Tetrahydroxy-4-methoxy-3',5-di-(3,3-dimethylallyl)-dihydrochalcone (**1**). Gum. IR ν_{\max} cm⁻¹: 3460, 2921, 1628, 1508, 1447, 1370, 1286, 1139, 1078, 994, 842. ¹H and ¹³C NMR (see Table 1). EIMS, *m/z* (rel. int.): 440 [M]⁺ (9), 438 (5), 246 (47), 218 (52), 205 (88), 203 (44), 177 (31), 165 (100), 137 (41), 85 (62), 83 (87), 69 (71).

2',3,6'-Trihydroxy-4-methoxy-5-(3,3-dimethylallyl)-3',4'-(2'',2''-dimethyldihydropyran)-dihydrochalcone (**2**). Gum. ¹H NMR, see Table 1. EIMS, *m/z* (rel. int.): 440 [M]⁺ (13), 438 (2), 246 (65), 220 (4), 218 (47), 205 (71), 203 (43), 193 (35), 177 (20), 165 (100), 137 (28), 69 (21).

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