

Article

Coumarins and Alkaloids from the Stems of *Metrodorea Flavida*

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Uma nova cumarina, a 5,6-metilenodioxo-7,8-dimetoxicumarina, foi isolada a partir dos galhos de *Metrodorea flavida*, juntamente com outras cumarinas e alcalóides conhecidos. As estruturas das substâncias isoladas foram definidas por análises de seus dados espectrais, bem como por comparação com dados da literatura.

A new coumarin, 5,6-methylenedioxy-7,8-dimethoxycoumarin has been isolated from the stems of *Metrodorea flavida*, as well as known coumarins and alkaloids. The structures of the new and the known compounds were established by spectral data and by comparison with the literature data.

Keywords: *Metrodorea flavida*, Rutaceae, coumarins, alkaloids

Introduction

In a previous paper, we reported the characterization of a new coumarin, 8-(2,3-dihydroxy-3-methylbutyloxy)-6,7-methylenedioxy coumarin, together with known furocoumarins and a furofuran lignan, which were isolated from the leaves of *Metrodorea flavida*¹. In continuation of our phytochemical studies on the constituents of this species, we report from the stem the isolation and structural elucidation of a new coumarin 5,6-methylenedioxy-7,8-dimethoxycoumarin (**1**), in addition to the known compounds: scoparone (**2**)²; 6,7-methylenedioxy-8-methoxycoumarin (**3**)³; xanthotoxin (**4**)⁴; isopimpinellin (**5**)⁴; imperatorin (**6**)⁴; braylin (**7**)⁵; γ -fagarine (**8**)⁶; kokusaginine (**9**)⁷; maculin (**10**)⁸; syringic aldehyde (**11**)⁹; rutaecarpine (**12**)¹⁰; sitosterol and lupeol.

All the compounds were isolated by chromatographic techniques. The structural elucidation of these compounds were based on spectrometric data, especially IR, ¹H NMR and ¹³C NMR, involving comparison with the literature data.

Experimental

Equipment

Mps uncorr. IR were recorded in KBr discs. ¹H and ¹³C-NMR spectra were recorded at 300 and 75 MHz, respectively, in CDCl₃ on a Varian GEMINI 300 instrument and at 400 and 100 MHz, in DMSO, on a Bruker

ARX 400 instrument. EIMS were obtained by direct probe insertion at 70 eV.

Plant material

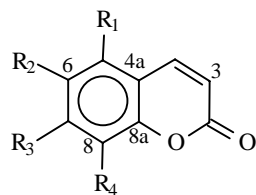
Metrodorea flavida was collected in Paragominas, State of Pará, Brazil, in December 1991. A voucher specimen is deposited at the Herbarium of the CPATU-EM-BRAPA, Belém, Brazil.

Extraction and isolation

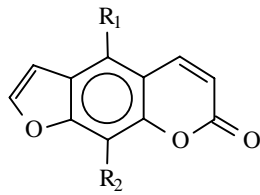
After drying, stems (231 g) were ground and percolated with hexane and CH₂Cl₂, successively. The concd. hexane extract (3.5 g) was submitted to CC using silica gel 60 Merck (particle size 0.063-0.200 mm) packed in hexane. Elution was performed with a gradient of hexane, Me₂CO and MeOH, affording 22 frs. The frs 3 and 7 after prep. TLC (silica gel and hexane-CH₂Cl₂-MeOH/10:10:0.1) yielded lupeol and sitosterol, respectively.

Fr. 12 was rechromatographed on silica gel using gradients of hexane, CH₂Cl₂ and MeOH. Those frs. containing homogeneous components, as judged by TLC, were combined and the solvent removed. Frs. 12/9-12 and 12/14-17 subjected to prep. TLC on silica gel (hexane-Me₂CO/75:25) afforded **6** (31.4 mg) and **12** (7.5 mg), respectively. Fr. 17 was subjected to CC on silica gel eluting with gradients of hexane, CH₂Cl₂, Me₂CO and MeOH affording **9** (3.1 mg).

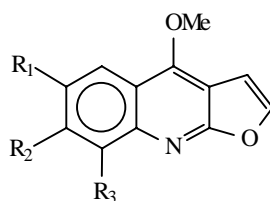
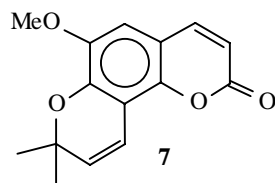
The CH₂Cl₂ extract (4.2 g) was subjected to chromatographic treatments similar to those used for the hexane



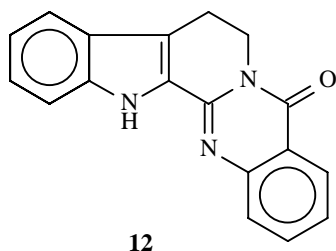
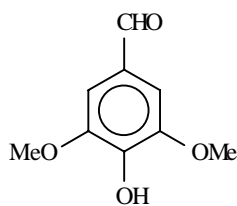
- 1** R₁-R₂ = OCH₂O,
R₃ = R₄ = OCH₃
1a R₁ = R₄ = OCH₃,
R₂-R₃ = OCH₂O
1b R₁ = R₂ = OCH₃,
R₃-R₄ = OCH₂O
2 R₁ = R₄ = H,
R₂ = R₃ = OCH₃
3 R₁ = H,
R₂-R₃ = OCH₂O,
R₄ = OMe



- 4** R₁ = H, R₂ = OCH₃
5 R₁ = R₂ = OCH₃
6 R₁ = H,
R₂ = OCH₂CH=C(CH₃)₂



- 7**
8 R₁ = R₂ = H, R₃ = OMe
9 R₁ = R₂ = OMe, R₃ = H
10 R₁-R₂ = OCH₂O, R₃ = H

**12****11**

extract. Frs. 4-5 afforded a mixture (6.3 mg) of lupeol and sitosterol. The Fr. 7 afforded **6** (4.8 mg). Fr. 8 was rechromatographed on silica gel using gradients of hexane, Me₂CO and MeOH yielding **4** (3.7 mg), **6** (8.1 mg), **7** (5.2 mg) and **12** (1.3 mg). Fr. 9 was subjected to CC on silica gel using hexane, Me₂CO and MeOH at different ratios of increasing polarity to give 7.0 mg of the new coumarin **1**, **2** (2.7 mg), **3** (0.5 mg), **4** (6.3 mg), **5** (4.3 mg), **8** (2.3 mg), **10** (3.2 mg), **11** (2.4 mg) and **12** (1.4 mg).

5,6-Methylenedioxy-7,8-dimethoxycoumarin (1). Amorphous solid. ¹H and ¹³C-NMR: see Table 1.

Results and Discussion

The new coumarin **1** was obtained from the dichloromethane extract of *M. flavida* and showed as a blue

Table 1. NMR data for compound **1** (CDCl₃, *J* values, in Hz, are given in parentheses).

Position	¹³ C	¹ H	COLOC
2	160.5		H-4
3	112.0	6.20, <i>d</i> (9.7)	
4	138.8	7.91, <i>d</i> (9.7)	
4a	107.0		H-3
5	142.9		OCH ₂ O
6	132.8		OCH ₂ O
7	133.4		MeO-7
8	126.9		MeO-8
8a	143.7		H-4
7-OMe	61.1	4.04, <i>s</i>	
8-OMe	61.1	3.99, <i>s</i>	
OCH ₂ O	102.1	6.01, <i>s</i>	

Hydrogens correlating with carbon resonance.

color on TLC under UV light 336 nm. Its ¹H NMR spectrum exhibited resonances typical of H-3 and H-4 (δ 6.20 and 7.91) of the coumarin nucleus in which C-5 was oxygenated¹¹. The presence of a methylenedioxy group was indicated from the methylene hydrogens signal at δ 6.01 (*s*) and two methoxy groups from the signals at δ 4.04 and 3.99 (*s*, 3H each one). These groups were located at the positions 5, 6, 7 and 8 suggesting one of the possible structures **1**, **1a** or **1b**. Further information concerning the actual positions of the methoxy groups was obtained by analysis of the 1D-NOE difference spectrum. Irradiation of the signal at δ 7.91 (*d*, H-4) didn't enhance the signals at δ 4.04 and 3.99 (2x OMe), which confirmed the placement of the methylenedioxy group at C-5/C-6. This result is consistent with the structure **1**. The assignments of the signals of carbons 3,4 and -OCH₂O- moiety in the ¹³C-NMR spectrum were established by HETCOR (¹³C, ¹H-COSY-¹J_{CH}) spectrum and the attribution of the quaternary carbons was made by COLOC (¹³C, ¹H-COSY-ⁿJ_{CH}, n = 2 and 3) experiment (Table 1).

This is the first report of the occurrence of one indolopiridoquinazoline alkaloid (rutaecarpine) from the genus *Metrodorea*. This fact reinforces the taxonomic position of the *Metrodorea flavida* into the Cusparieae tribe, due to the occurrence of this type of alkaloid in the tribes Cusparieae and Xanthoxyleae.

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References

1. Baetas, A.C.; Arruda, M.S.P.; Müller, A.H.; Arruda, A.C. *Phytochemistry* **1996**, *43*, 491.
2. Adegosam, E.K. *Phytochemistry* **1973**, *12*, 2310.
3. Sarker, S.D.; Gray, A.I.; Waterman, P.G. *J. Nat. Prod.* **1994**, *57*, 1549.
4. Razdam, T.K.; Qadri, B.; Harkar, S.; Waight, E. *Phytochemistry* **1987**, *27*, 2063.
5. Silva, M.; Cruz, M.A. *Phytochemistry* **1971**, *10*, 3255.
6. Tillequin, F.; Koch, M.; Sevenet, T.H. *Planta Medica* **1980**, *38*, 3830.
7. Khalid, S.A.; Waterman, P.G. *Phytochemistry* **1981**, *20*, 2761.
8. Dreyer, D.L. *Phytochemistry* **1980**, *19*, 941.
9. Pouchert, C.J. In *The Aldrich Library of NMR Spectra*, 2nd ed, Aldrich Chemical Company Inc., USA, 1983, vol. 2, p 179.
10. Breitmaier, E.; Voelter, W. In *Carbon-13 NMR Spectroscopy*, 3rd ed., UCH, Weinheim, 1987, p 447.
11. Murray, R.D.H.; Mendez, J.; Brown, S.A. In *The Natural Coumarins: Occurrence, Chemistry and Biochemistry*, John Wiley & Sons Ltd, New York, 1982, p 21.

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