Pergamon

PII: S0031-9422(96)00447-5

Phytochemistry, Vol. 44, No. 2, pp. 347–349, 1997

0031-9422/97 $17.00 + 0.00

TWO FLAVANS FROM *BROSIMUM ACUTIFOLIUM**

SERGIO LUIZ TORRES, JOÃO CARLOS M. MONTEIRO, MARA SILVIA P. ARRUDA,† ADOLFO H. MÜLLER and ALBERTO C. ARRUDA

Laboratório de Química, Universidade Federal do Pará, Campus Universitário, 66075–900 Belém, Brazil

(*Received* 12 *April* 1996)

Key Word Index—*Brosimum acutifolium*; Moraceae; bark; new flavans.

Abstract—Two new flavans were isolated from a dichloromethane extract of the bark of *Brosimum acutifolium* and identified as 4′-hydroxy-7,8-(2″,2″-dimethylpyran)flavan and 4′-hydroxy-7,8-(3″-hydroxy-2″,2″-dimethylpyran)flavan. Structures were elucidated by spectral data. Copyright © 1996 Elsevier Science Ltd

INTRODUCTION

The genus *Brosimum* is considered to contain *ca* 13 species that occur in tropical America [1]. To date, only two species have been examined phytochemically, *B. gaudichaudii* Trecul, the roots of which contain furocoumarins [2], and *B. rubescens* Taubert, the trunk wood of which contains coumarins and pyranocoumarins [3]. The remaining 11 species were compared with those only by TLC analysis [4].

Brosimum acutifolium subsp. *acutifolium*, popular name mururé, has been used in folk medicine in the Amazon region as an anti-inflammatory and anti-rheumatic agent [5]. The present work describes the isolation and identification of two new flavans from a sample of bark of this species.

RESULTS AND DISCUSSION

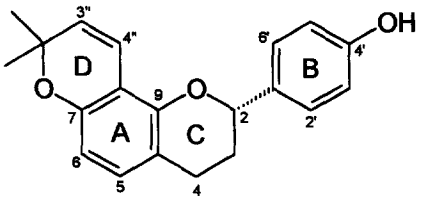
The dichloromethane extract of *B. acutifolium* was fractionated as described in the Experimental section and yielded the flavans **1** and **2**.

Determination of the M_r by EI-mass spectrometry (*m/z* 308, $[M]^+$) allied to hydrogen, carbon and hydroxyl counts by NMR, revealed the formula $C_{20}H_{19}O_2$ (OH) for compound **1**. The location of the hydroxyl group at C-4′ was readily apparent from the detection of an AA′ BB′-system [(δ 7.29 (2H, *d*, $J = 8.5$ Hz, H-2′, 6′); δ 6.86 (2H, *d*, $J = 8.5$ Hz, H-3′, 5′)] in ^{1}H NMR spectrum, supported by the peak at *m/z* 120 in the EI-mass spectrum (retro Diels–Alder styryl fragment), suggesting the presence of a *para*-substituted B-ring. This spectrum also showed two *ortho*-coupled doublets ($J = 8.2$ Hz), each one integrating for one proton at δ 6.82 and 6.35, assigned to the H-5 and H-6 protons, respectively. The five aliphatic protons in the C-ring were observed as one double-doublet at δ 4.98 ($J = 10.9$ and 2.2 Hz) assigned to the H-2 proton, two multiplets each one integrating for one proton, centred at δ 2.02 and 2.15, attributed to the H-3 protons and two signals at δ 2.89 (1H, *ddd*, $J = 16.3$; 11.2 and 5.6 Hz) and 2.69 (1H, *ddd*, $J = 16.3$; 5.3 and 3.4 Hz) attributed to H-4β and H-4α, respectively. The fragment ion at *m/z* 293 ([M-Me]$^+$, 17.6%) indicated the presence of a 2,2-dimethylchromene ring, which was confirmed by the sharp singlet of six protons intensity at δ 1.41 assigned to a gem-dimethyl group, which is adjacent to an oxygen atom and two doublets, each having one proton intensity and $J = 9.9$ Hz, observed at δ 5.51 and 6.67 assigned to H-3″ and H-4″, respectively. A NOESY experiment revealed correlation between H-5 and H-4α, establishing the position of the 2,2-dimethylchromene ring as depicted in the structure of compound **1**. The ^{13}C NMR spectrum was consistent with a structure of flavan compounds [6] (Table 2). It showed 13 signals attributed to 15 carbons of the flavan skeleton, beyond four signals assigned to the five carbons of the 2,2-dimethylchromene ring. The assignments of the carbon signals were confirmed by a DEPT pulse sequence (Table 2) and HETCOR experiment (Table 1).

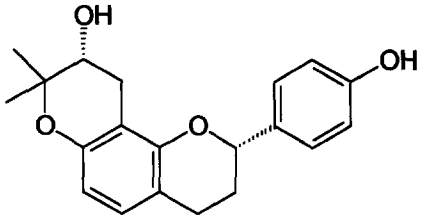
Compound **2** had the molecular formula $C_{20}H_{18}O_2(OH)_2$, with 18 μa (H_2O) more than compound **1**. As shown in Table 1, it exhibited a pattern of signals in the ^{1}H NMR spectrum similar to that of compound **1**, except for the absence of *cis*-olefinic protons and instead the presence of a triplet at δ 3.77 (1H, $J = 5.2$ Hz) characteristic of a carbonolic proton, attributed to H-3″, this was confirmed by a heteronuclear multiple bond correlation (COLOC) experiment (Table 1), optimized for $^2J_{(CH)}$ and $^3J_{(CH)}$ coupling, which revealed $^3J_{(CH)}$ coupling between the methyl protons in C-2″ and carbon 3″. Overlap in the ^{1}H NMR

*This paper is based in part on the M.Sc. dissertation that will be presented by S.L.T. to the Universidade Federal do Pará, Belém-Pará, Brazil.

†Author to whom correspondence should be addressed.



1



2

spectrum of the signals of H-4 and H-4″ at δ 2.71 and 2.90, each one integrating for two protons, did not permit the establishment of the precise position of C-5/C-6 or C-7/C-8 of the 2,2-dimethylchromene ring by a NOE differential experiment. The position of the 2,2-dimethylcromene ring was defined by a COLOC experiment from the observation of $^3J_{(CH)}$ coupling between the H-5 and C-4, confirming the angular orientation at C-7 and C-8. The ^{13}C NMR spectrum was assigned by DEPT pulse sequence (Table 2) and a HETCOR experiment (Table 1). To the carbons C-4 and C-4″ were attributed the signals at δ 25.0 and 26.8, respectively, through the COLOC experiment which revealed $^3J_{(CH)}$ coupling between C-4 and H-2 beyond C-4 and H-5.

EXPERIMENTAL

General. Mps uncorr. ^{1}H NMR were recorded at 300 and 400 MHz in $CDCl_3$ and ^{13}C NMR at 75 MHz. EI-MS were obtained by direct probe insertion at 70 eV. IR were recorded in NaCl film.

Plant material. Brosimum acutifolium was collected in Portel county, State of Pará, Brazil, and identified by Dr Maria Elisabeth van den Berg of the Botanic Department of Museu Paraense Emílio Goeldi, Belém, Pará.

Extraction and isolation. Powdered bark (2.3 kg) was extracted with hexane, CH_2Cl_2, EtOAc and MeOH, successively. After evapn of solvent the CH_2Cl_2 extract (4 g) was subjected to CC on silica gel (70–230 mesh) and eluted with hexane, CH_2Cl_2 and MeOH in increasing order of polarity, furnishing 40 frs. The frs were analysed by silica gel TLC using hexane–CH_2Cl_2–MeOH (10:10:1). Compounds were detected by UV and spraying with 10% alcoholic H_2SO_4 or ceric sulphate.

Frs 18–22 were rechromatographed on silica gel and eluted with hexane and a gradient of Me_2CO to yield 44.5 mg of compound **1**. Frs 33–34 after three purifications on silica gel using different ratios of increasing polarity of hexane–Me_2CO, hexane–CH_2Cl_2 and hexane–CH_2Cl_2, successively, yielded compound **2** (12.0 mg).

4′-Hydroxy-7,8-(2″, 2″-dimethylpyran)flavan (**1**). Yellow gum. $[\alpha]_D$ −34.9° ($CHCl_3$ *c* 1.2). $IR_{max}cm^{-1}$: 3401, 2921, 1607, 1516, 1473. $\lambda_{max}^{CHCl_3}$ nm: 260 (0.643), 282 (0.943), 294 (0.750), 310 (0.321). ^{1}H NMR: Table 1. ^{13}C NMR: Table 2. EI-MS *m/z* (rel. int.): 308 $[M]^+$ (9), 293 $[M-Me]^+$ (17.6), 173 $[C_{11}H_9O_2]^+$ (31.4), 120 $[C_8H_8O]^+$ (19.2), 107 $[C_7H_7O]^+$ (10.2), 55 $[C_4H_7]^+$ (100).

4′-Hydroxy-7,8-(3″-hydroxy-2″,2″-dimethylpyran)-flavan (**2**). Gum. $[\alpha]_D$ −14.4° ($CHCl_3$, *c* 1.7). IR ν_{max} cm^{-1}: 3363, 3010, 1614, 1594, 1516, 1484, 1446. $\lambda_{max}^{CHCl_3}$ nm; 281 (1.162). ^{1}H NMR: Table 1. ^{13}C NMR: Table 2. EI-MS *m/z* (rel. int.): 326 $[M]^+$ (93.6), 308 $[M-H_2O]^+$ (4), 293 $[308-Me]^+$ (14), 173 $[C_{11}H_9O_2]^+$ (13), 120 $[C_8H_8O]^+$ (100), 107 $[C_7H_7O]^+$ (51).

Table 1. ^{1}H NMR, HETCOR, NOESY and COLOC data for compounds **1** and **2**

	1			**2**		
Position	^{1}H NMR (δ)	HETCOR (^1J ^{13}C–^{1}H)	NOESY (^{1}H–^{1}H)	^{1}H NMR (δ)	HETCOR (^1J ^{13}C–H)	COLOC (^2J,^3J ^{13}C–H)
2	4.98 (*dd*, *J* = 10.9, 2.2 Hz)	77.2	H-3	4.96 (*dd*, *J* = 11.4, 2.2 Hz)	77.6	C-4, C-2′, C-6′
2H-3	2.00–2.04 (*m*)	30.0	H-4, H-2,	2.14–2.18 (*m*)	30.2	–
	2.13–2.17 (*m*)		H-2′, H-6′	1.98–2.02 (*m*)		
1H-4β	2.86 (*ddd*, *J* = 16.3, 11.2, 5.6 Hz)	24.7	H-3	2.86–2.94 (*m*)	25.0	C-10
1H-4α	2.69 (*ddd*, *J* = 16.3, 5.3, 3.4 Hz)	24.7	H-3, H-5	2.68–2.74 (*m*)	25.0	C-10
5	6.82 (*d*, *J* = 8.2 Hz)	128.9	H-4, H-6	6.85 (*d*, *J* = 8.3 Hz)	128.0	C-4, C-7, C-9
6	6.35 (*d*, *J* = 8.2 Hz)	108.7	H-5	6.41 (*d*, *J* = 8.3 Hz)	109.4	C-10
2H-2′, 6′	7.29 (*d*, *J* = 8.5 Hz)	127.4	H-3	7.27 (*d*, *J* = 8.3 Hz)	127.6	C-2, C-4′
2H-3′, 5′	6.86 (*d*, *J* = 8.5 Hz)	115.3		6.82 (*d*, *J* = 8.3 Hz)	115.5	C-2′, C-6′, C-1′
3″	5.51 (*d*, *J* = 9.9 Hz)	128.8	H-4″	3.77 (*t*, *J* = 5.2 Hz)	69.8	–
4″	6.67 (*d*, *J* = 9.9 Hz)	117.0	H-3″	–	–	–
2H-4″	–	–	–	2.86–2.94 (*m*)	26.8	C-8
				2.68–2.74 (*m*)		
2 Me	1.41 (*s*)	27.8		1.35 (*s*)	24.7	C-2, C-3″
				1.30 (*s*)	22.4	

Solution in $CDCl_3$ referenced to $CHCl_3$ at δ 7.26 (^{1}H) and δ 77.23 (^{13}C).

Table 2. ^{13}C NMR spectral data of compounds **1** and **2**

Position	1		2	
	^{13}C NMR	DEPT	^{13}C NMR	DEPT
2	77.2	CH	77.6	CH
3	30.0	CH_2	30.2	CH_2
4	24.7	CH_2	25.0	CH_2
5	128.9	CH	128.0	CH
6	108.7	CH	109.4	CH
7	151.9*	C	153.6*	C
8	110.0	C	107.6	C
9	150.4*	C	151.9*	C
10	113.9	C	113.4	C
1′	134.3	C	134.4	C
2′	127.4	CH	127.6	CH
3′	115.3	CH	115.5	CH
4′	155.1	C	155.5	C
5′	115.3	CH	115.5	CH
6′	127.4	CH	127.6	CH
2″	75.6	C	76.6	C
3″	128.8	CH	69.8	CH
4″	117.0	CH	26.8	CH_2
2Me	27.8	CH_3	24.7	CH_3
			22.3	

Solution in $CDCl_3$ referenced to $CHCl_3$ at δ 7.26 (^1H) and δ 77.23 (^{13}C).
*Interchangeable signals.

Acknowledgements—The authors are grateful to Coodenação de Aperfeiçoamento de Pessoal de Ensino Superior (CAPES) for financial support, to Universidade Federal de São Carlos (São Carlos–São Paulo–Brazil) for ^1H NMR spectra at 400 MHz and to Museu Paraense Emilio Goeldi, Belém, PA, Brazil, for GC/MS analysis. NMR studies (300 and 75 MHz) were performed in the Universidade Federal do Pará NMR Laboratory.

REFERENCES

1. Allaby, M. (1992) *The Concise Oxford Dictionary of Botany*, p. 58. Oxford University Press, Oxford.
2. Pozetti, G. L. (1969) *Rev. Fac. Farm. Odont. Araraquara Brasil* **3**, 215.
3. Filho, R. Braz, Magalhães, A. F. and Gottlieb, O. R. (1972) *Phytochemistry* **11**, 3307.
4. Gottlieb, O. R., Silva, M. L. and Maia, J. G. S. (1972) *Phytochemistry* **11**, 3479.
5. Berg, M. E. van den (1982) *Plantas Medicinais da Amazônia. Contribuição ao Seu Conhecimento Sistemático*, 2nd edn, p. 97. Belém, CNPQ/PTU/MPEG.
6. Saini, K. S. and Ghosal, S. (1984) *Phytochemistry* **23**, 2415.