Phytochemistry, 1972, Vol. 11, pp. 3515 to 3519. Pergamon Press. Printed in England.

FLAVONOIDS AND COUMARINS FROM *PLATYMISCIUM PRAECOX**

A. BRAGA DE OLIVEIRA, L. G. FONSECA E SILVA and O. R. GOTTLIEB
Instituto de Ciências Exatas, Universidade Federal de Minas Gerais, Belo Horizonte, Brasil

(*Received* 21 *June* 1972. *Accepted* 10 *June* 1972)

Key Word Index—*Platymiscium praecox*; Leguminosae; isoflavone; flavanones; isoliquiritigenin; flavones; new C-methylcoumarins.

Abstract—The wood of *Platymiscium praecox* Mart. (Leguminosae-Lotoideae) contains sitosterol, 4,2′,4′-trihydroxychalcone, (2*R*)-7-hydroxyflavanone, (±)-7,4′-dihydroxyflavanone, (2*S*, 3*S*)-3,7-dihydroxyflavanone, 3,7-dihydroxyflavone, 3,7,4′-trihydroxyflavone, 6,7-dihydroxy-4′-methoxyisoflavone and 6,7-dimethoxycoumarin. It also contains three novel compounds: 7-hydroxy-4-methoxy-5-methylcoumarin, 7-*O*-glucosyloxy-4-methoxy-5-methylcoumarin and 7-hydroxy-4,8-dimethoxy-5-methylcoumarin.

Platymiscium praecox Mart. (Leguminosae-Lotoideae) is an arboreous species which occurs near Pedro Leopoldo, Minas Gerais State, Brazil. Its trunkwood yielded a series of crystalline compounds which were classified by their spectra as a chalcone, three flavanones, two flavones, an isoflavone and four coumarins.

The chalcone and one of the flavanones were identified with the well known 4,2′,4′-trihydroxychalcone (isoliquiritigenin) and (±)-7,4′-dihydroxyflavanone (liquiritigenin)[3] through direct comparison with synthetic samples.[4,5] The spectral characterization of the second flavanone as (2*R*)-7-hydroxyflavanone was consubstantiated by direct comparison with synthetic (±)-7-hydroxyflavanone.[6-8]

The PMR spectrum of the third flavanone included the pair of doublets typical of the AB system of vicinal protons at positions 2 and 3 of 3-hydroxyflavanones.[9] The B part of this signal was shifted paramagnetically by 1·2 ppm upon acetylation of the compound, confirming the existence of a secondary carbinol. The remaining features of the PMR spectrum defined the substitution pattern of the aromatic rings and, in conjunction with

* Part XXXVIII in the series "The Chemistry of Brazilian Leguminosae". For Part XXXVII see Ref. 1. For preliminary communication see Ref. 2. Taken from the Doctorate-thesis submitted by Luiz Gonzaga Fonseca e Silva to the Universidade Federal de Minas Gerais (1972).

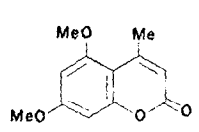
[1] O. R. GOTTLIEB and J. RÊGO DE SOUSA, *Phytochem.* **11**, 2841 (1972).
[2] A. BRAGA DE OLIVEIRA, M. DE L. CARVALHO DANTAS MOREIRA, L. G. FONSECA E SILVA, O. R. GOTTLIEB and C. RIBEIRO DE CASTRO, *Anais Acad. Brasil. Ciênc.* **42** (Supplement), 109 (1970).
[3] M. SHIMOKORIYAMA, in *The Chemistry of Flavonoid Compounds* (edited by T. A. GEISSMAN), p. 286, Pergamon Press, Oxford (1962).
[4] E. C. BATE-SMITH and T. SWAIN, *J. Chem. Soc.* 2185 (1953).
[5] S. HISHIDA, *Nippon Kagaku Zasshi* **76**, 204 (1955); *Chem. Abs.* **51**, 17901 (1957).
[6] D. S. MITTAL, *Proc. Natl. Acad. Sci. India* **15A**, 8 (1946); *Chem. Abs.* **44**, 3941 (1950).
[7] R. BOGNAR, I. FARKAS and M. RAKOSI, *Acta Chim. Acad. Sci. Hung.* **30**, 87 (1962); *Chem. Abs.* **58**, 4635 (1963).
[8] L. PONNIAH and T. R. SESHADRI, *Proc. Indian Acad. Sci.* **37A**, 534 (1953); *Chem. Abs.* **48**, 11402 (1954).
[9] L. M. JACKMAN, in *Progress in the Chemistry of Organic Natural Products* (edited by L. ZECHMEISTER), Vol. XXIII, p. 315, Springer, Wien (1965).

MS and ORD data,[10] led to the structure of (2*S*, 3*S*)-3,7-dihydroxyflavanone. Only synthetic (±)-3,7-dihydroxyflavanone has been described previously.[11]

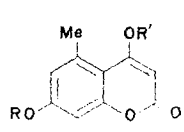
The flavones were identified as 3,7-dihydroxyflavone and 3,7,4′-trihydroxyflavone by spectral means. The data obtained for these isolates and for the derived acetates are in agreement with the data reported for synthetic 3,7-dihydroxyflavone,[12,13] as well as for synthetic[14] and natural[15] 3,7,4′-trihydroxyflavone.

The PMR spectrum of its diacetate identified the isoflavone unequivocally with 6,7-dihydroxy-4′-methoxyisoflavone (texasin), previously detected in *Baptisia australis* by PC.[16] The isolate has a m.p. and gave UV and IR spectra as required by the literature for synthetic specimens.[17–19]

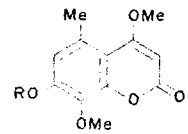
One of the four coumarins of *P. praecox* was identified with 6,7-dimethoxycoumarin (aesculetin dimethyl ether) isolated previously from plants.[20,21] The remaining three compounds were immediately recognized as unusual: their PMR spectra indicated in each case the presence of a *C*-methyl group. This fact, combined with elementary, mass spectral and functional analyses, led to the formula $C_9H_3O_2.CH_3.OH.OCH_3$ for one of them. The hydroxyl must be placed at a position such as C-7, conjugated with the carbonyl, since upon methylation and upon acetylation a shift of the IR carbonyl stretching band from 1700 to 1725 cm^{-1} was observed. Two of the three undefined hydrogens in the formula must be *meta*-related and both vicinal to the hydroxyl. In the acetate they give rise to a pair of doublets (*J* 2·5 Hz) at a significantly lower field (τ 3·10 and 3·25) than in the methyl ether (2 proton singlet, τ 3·40). The third hydrogen was placed at the 3 position of the heterocycle, in view of the relatively small chemical shift of its PMR signal [*s*, τ 4·50 (methyl ether), 4·40 (acetate)]. At this point two structural alternatives (I and IIb) had to be considered for the methyl ether. A synthetic sample of 5,7-dimethoxy-4-methylcoumarin (I),



(I)



	R	R′
(IIa)	H	Me
(IIb)	Me	Me
(IIc)	COMe	Me
(IId)	Me	H
(IIe)	glucosyl	Me
(IIf)	tetra-acetyl-glucosyl	Me
(IIg)	glucosyl	H
(IIh)	tetra-acetyl-glucosyl	COMe



	R
(IIIa)	H
(IIIb)	Me
(IIIc)	COMe

however, proved to be different, upon direct comparison, with this derivative. Most significantly, the PMR signal due to the *C*-methyl group of the synthetic compound appeared as a doublet (τ 7·48, *J* 1·5 Hz) and the vicinal olefinic proton gave a quartet (τ 4·06, *J* 1·5 Hz), as in other coumarins and chromones of this type. In contradistinction, methyl substituted aromatics with a free *ortho*-position, usually show the methyl proton signal as a

[10] W. GAFFIELD and A. C. WAISS, JR., *Chem. Commun.* 29 (1968).
[11] T. OYAMADA and H. BABA, *Nippon Kagaku Zasshi* **86**, 750 (1965).
[12] T. HEAP and R. ROBINSON, *J. Chem. Soc.* 2336 (1926).
[13] V. DEULOFEU and N. SCHOPFLOCHER, *Gazz. Chim. Ital.* **83**, 449 (1953).
[14] Z. I. JERZMANOWSKA and M. MICHALSKA, *Roczniki Chem.* **35**, 353 (1961); *Chem. Abs.* **55**, 18715 (1961).
[15] E. WONG, P. I. MORTIMER and T. A. GEISSMAN, *Phytochem.* **4**, 89 (1965).
[16] K. R. MARKHAM, W. T. SWIFT and T. J. MABRY, *J. Org. Chem.* **33**, 462 (1968).
[17] J. B. HARBORNE, O. R. GOTTLIEB and M. TAVEIRA MAGALHÃES, *J. Org. Chem.* **28**, 881 (1963).
[18] S. F. DYKE, W. D. OLLIS, M. SAINSBURY and J. S. P. SCHWARZ, *Tetrahedron* **20**, 1331 (1964).
[19] K. FUKUI and T. MATSUMOTO, *J. Sci. Hiroshima Univ.* Ser. A II, **28** (1), 47 (1964); *Chem. Abs.* **62**, 9097 (1965).
[20] F. E. KING, J. R. HOUSLEY and T. J. KING, *J. Chem. Soc.* 1392 (1954).
[21] T. OHTA, *J. Pharm. Soc. Japan* **66**, 11 (1946).

somewhat widened singlet, an observation extensive to the spectra of the methyl ether and the acetate of our natural product. This must then possess the structure of 7-hydroxy-4-methoxy-5-methylcoumarin (IIa). The entire analysis, however, had relied upon an original postulate which formulated the compound as a coumarin. It was thus deemed desirable to confirm the proposal by degradative and synthetic studies.

Initially it was ascertained that vigourous alkaline treatment of the isolate does lead, as expected,[22] to 2,4-dihydroxy-6-methylacetophenone, or, under methylating conditions, to 2,4-dimethoxy-6-methylacetophenone. The acetophenones, obtained in good yield, were identified by direct comparison with synthetic samples.[23,24] The substitution of the aromatic ring thus having been confirmed, our attention was directed towards the heterocycle. 4-Methoxycoumarins are easily demethylated upon reflux with 18% aq. HCl for short periods.[25] Indeed, when the methyl ether of the natural compound was submitted to this procedure, 4-hydroxy-7-methoxy-5-methylcoumarin resulted. This was characterized unequivocally by direct comparison with a synthetic sample, obtained from 2-hydroxy-4-methoxy-6-methylacetophenone[23] and diethyl carbonate in the presence of sodium,[26] and was converted to the methyl ether of compound IIa by treatment with dimethyl sulphate. The structure of 7-*O*-glucosyloxy-4-methoxy-5-methylcoumarin (IIe) was attributed to the third coumarin, when it was found that its acid hydrolysis leads to glucose and IIa.

To the fourth coumarin we assign tentatively the structure of 7-hydroxy-4,8-dimethoxy-5-methylcoumarin (IIIa). The PMR spectra of IIIa and IIa are very similar, inclusively with respect to indications of benzylic coupling involving an aromatic proton and the *C*-methyl protons. The significant difference refers to the presence of a sole aromatic proton signal in the spectrum of IIIa (a singlet at τ 3·31), the additional aromatic proton signal being replaced by a methoxyl singlet (τ 6·10). An alkaline degradation was performed on the small quantity available. The amount of acetophenone (positive $FeCl_3$-test) obtained, however, was too small for full characterization.

EXPERIMENTAL

M.ps. were taken on the Kofler block and are uncorrected. Column chromatography employed Merck's Kieselgel 0·05–0·20 mm. PMR spectra were taken on a Varian HA-60-IL instrument. TMS was used as internal standard, *s*—singlet, *d*—doublet, *dd*—double doublet, *m*—multiplet. MS were recorded with an AEI MS9 instrument. ORD curves were recorded with a Cary 60 spectropolarimeter. The acetates were obtained with Ac_2O-pyridine at room temp. Unless otherwise stated, the methyl ethers were obtained with Me_2SO_4–K_2CO_3–acetone under reflux.

Isolation of the constituents of Platymiscium praecox. Softwood (7 kg) and heartwood (10 kg) were extracted successively with benzene and with EtOH. A portion (25 g) of the benzene extract (48 g) of the softwood was chromatographed on silica (600 g), yielding the following useful fractions with the indicated eluants: A_1 (benzene), A_2 and A_3 ($CHCl_3$–MeOH, 97·5:2·5). A_1 was recrystallized from EtOH–H_2O (1:1) giving sitosterol (90 mg). A_2 and A_3 were recrystallized from toluene–AcOH (1:1) giving IIa (210 mg) and IIIa (169 mg). A portion (15 g) of the EtOH extract (180 g) of the softwood was chromatographed on silica (450 g), yielding the following useful fractions with the indicated eluants: B_1 ($CHCl_3$–MeOH, 9:1), B_2 ($CHCl_3$–MeOH, 4:1). B_1 was recrystallized from toluene–AcOH (1:1) giving IIa (189 mg). B_2 was recrystallized from MeOH giving IIe (128 mg).

Upon partial concentration and cooling of the benzene solution obtained by extraction of the heartwood, a solid (28 g) precipitated. This was separated by filtration and chromatographed on silica (450 g), yielding the following useful fractions with the indicated solvents: C_1–C_6 in order ($CHCl_3$), C_7 ($CHCl_3$–MeOH,

[22] G. Büchi, D. H. Klaubert, R. C. Shank, S. M. Weinreb and G. N. Wogan, *J. Org. Chem.* **36**, 1143 (1971).
[23] K. Hoesch, *Chem. Ber.* **48**, 1122 (1915).
[24] J. Tambor, *Chem. Ber.* **41**, 794 (1908).
[25] N. J. Desai and S. Sethna, *J. Org. Chem.* **22**, 388 (1957).
[26] J. Boyd and A. Robertson, *J. Chem. Soc.* 174 (1948).

19:1). C_1 was recrystallized from $CHCl_3$ giving 7-*hydroxyflavanone* (192 mg). C_2 was washed with Et_2O and recrystallized from toluene–AcOH (1:1) giving 3,7-*dihydroxyflavanone* (92 mg). C_3 was recrystallized from toluene–AcOH (1:1) giving 3,7-*dihydroxyflavone* (110 mg). C_4 was recrystallized from toluene–AcOH (1:1) giving IIa (90 mg). C_5 was recrystallized from EtOH–H_2O (1:1) giving 4,2′,4′-*trihydroxychalcone* (70 mg). C_6 was recrystallized from EtOH–H_2O (1:1) giving 7,4′-*dihydroxyflavanone* (350 mg). C_7 was recrystallized from toluene–AcOH (1:1) giving 3,7,4′-*trihydroxyflavone* (82 mg). The filtered benzene solution was evaporated. A portion (25 g) of the residue (109 g) was chromatographed on silica (550 g) yielding various fractions which after recrystallization gave 6,7-*dimethoxycoumarin* (350 mg), 7-*hydroxyflavanone* (500 mg), 3,7-*dihydroxyflavanone* (338 mg), 3,7-*dihydroxyflavone* (677 mg), 4,2′,4′-*trihydroxy chalcone* (898 mg) and 7,4′-*dihydroxyflavanone* (120 mg). A portion (50 g) of the EtOH extract (670 g) of the heartwood was chromatographed on silica (1 kg) yielding one useful fraction upon elution with $CHCl_3$–MeOH (97:3). This was purified by passage through Sephadex LH20 (MeOH) giving 6,7-*dihydroxy*-4′-*methoxyisoflavone* (44 mg).

4,2′,4′-*Trihydroxychalcone*. Yellow crystals, m.p. and m.m.p. with a synthetic sample 200–202° [lit.[4] m.p. 202–203°]. M found and required: 256.

(2R)-7-*Hydroxyflavanone*. Needles, m.p. and m.m.p, with a synthetic sample 190–191° [lit.[6,7] m.p. 190–191°[. ORD (EtOH, *c* 0·2, 350–275 nm): $[\phi]_{350}$ + 1200, $[\phi]_{342}$ + 2400, $[\phi]_{333}$ O, $[\phi]_{325}$ −8400, $[\phi]_{310}$ −21 600, $[\phi]_{298}$ −10 800, $[\phi]_{285}$ O, $[\phi]_{275}$ +4800. *Acetate*. M.p and m.m.p. with a synthetic sample 93–94° [lit[8] m.p. 104–105°].

(±)-7,4′-*Dihydroxyflavanone*. M.p. 197–198° [lit.[5] m.p. 196–197°]. UV identical to spectrum given in lit.[27] M found and required: 256. ORD curve close to base line.

(2S, 3S)-3,7-*Dihydroxyflavanone*. Needles, m.p. 155–158° [lit.[11] m.p. for a synthetic racemic sample 170–171°]. ν_{max}^{KBr} (cm^{-1}): 3430, 3240, 1655, 1615, 1570, 1465, 1145, 1110, 1000, 810. λ_{max}^{EtOH} (nm): 232, 278, 314 (ε 10 000, 12 500, 7700); $\lambda_{max}^{EtOH+NaOH}$ (nm): 257, 338 (ε 7000, 26 600); $\lambda_{max}^{EtOH+NaOAc}$ (nm): 258, 338 (ε 6600, 23 800). NMR [$(CD_3)_2CO$, τ]: 2·28 (*d*, *J* 8·0 Hz, H-5), 2·40–2·71 (m, $C_6\underline{H}_5$), 3·40 (*dd*, *J* 8·0 and 2·0 Hz, H-6), 3·60 (*d*, *J* 2·0 Hz, H-8), 4·87 (*d*, *J* 12·0 Hz, H-2), 5·47 (*d*, *J* 12·0 Hz, H-3). MS: M 256 (25%), *m/e* (%) 227 (100), 163 (18), 149 (58), 137 (94), 136 (31), 121 (15), 120 (55), 108 (18), 91 (86). ORD (EtOH, *c* 0·2, 370–275 nm): $[\phi]_{370}$ +2560, $[\phi]_{345}$ +6400, $[\phi]_{335}$ O, $[\phi]_{330}$ −11 520, $[\phi]_{318}$ −32 000, $[\phi]_{308}$ −11 520, $[\phi]_{300}$ O, $[\phi]_{285}$ 21 760 $[\phi]_{275}$ +17 920. *Diacetate*. Needles, m.p. 91–92° ν_{max}^{KBr} (cm^{-1}): 1780, 1750, 1710, 1620, 1585, 1445, 1030, 910, 760, 700. RMN (CCl_4, τ): 2·13 (*d*, *J* 8·0 Hz, H-5), 2·63 (*s*, $C_6\underline{H}_5$), 3·24 (*dd*, *J* 8·0 and 2·0 Hz, H-6), 3·27 (*d*, *J* 2·0 Hz, H-8), 4·30 (*d*, *J* 12·0 Hz, H-3), 4·68 (*d*, *J* 12·0 Hz, H-2), 7·74 (*s*, ArOCOC$\underline{H}_3$), 8·02 (*s*, *R*OCOC$\underline{H}_3$).

3,7-*Dihydroxyflavone*. Light yellow crystals, m.p. 255–256° (MeOH) [lit. m.p. 258°,[12] 255–256°[13]]. ν_{max}^{KBr} (cm^{-1}): 3360, 1625, 1575, 1290, 1180, 770. λ_{max}^{EtOH} (nm): 227, 254, 325 inf., 345 (ε 15 700, 13 700, 15 100, 16600); $\lambda_{max}^{EtOH+NaOH}$ (nm): 244, 284, 341, 408 (ε 15 800, 13 500, 9200, 15 700). MS: M 254 (100%), *m/e* (%) 226 (11), 197 (17), 181 (5), 137 (5), 105 (14). *Diacetate*. Crystals, m.p. 157–159° (EtOH) [lit.[12,13] m.p. 157·5–158·5°]. ν_{max}^{KBr} (cm^{-1}): 1770, 1660, 1620, 1450, 1375, 1020, 965, 900, 770. NMR ($CDCl_3$, τ): 1·70 (*d*, *J* 8·5 Hz, H-5), 2·00–2·25 (*m*, H-2′, H-6′), 2·36–2·61 (*m*, H-3′, H-4′, H-5′), 2·74 (*d*, *J* 3·0 Hz, H-8), 2·82 (*dd*, *J* 8·5 and 3·0 Hz, H-6), 7·66 (*s*, two OCOC$\underline{H}_3$).

3,7,4′-*Trihydroxyflavone*. Light yellow crystals, m.p. 296–299° [lit.[14] m.p. 299–302°]. ν_{max}^{KBr} (cm^{-1}): 3480, 3390, 1625, 1450, 1275, 1180, 840. *Triacetate*. M.p. 243–247° (EtOH). ν_{max}^{KBr} (cm^{-1}): 1775, 1670, 1630, 1450, 1380, 1220, 1030. NMR ($CDCl_3$, τ): 1·70 (*d*, *J* 8·5 Hz, H-5), 2·07 (*d*, *J* 8·5 Hz, H-2′, H-6′), 2·59 (*d*, *J* 2·0 Hz, H-8), 2·71 (*d*, *J* 8·5 Hz, H-3′, H-5′), 2·81 (*dd*, *J* 8·5 and 2·0 Hz, H-6), 7·66 (*s*, three OCOC$\underline{H}_3$).

6,7-*Dihydroxy*-4′-*methoxyisoflavone*. M.p. 275–278° [lit. m.p. 278–280°,[19] 291·5–292·5 dec.[18]]. IR and UV data as required by lit.[17,18] MS: M 284 (100%), *m/e* (%) 269 (16), 231 (24), 165 (12), 152 (22), 132 (29). *Diacetate*. M.p. 175–178° (EtOH). ν_{max}^{KBr} (cm^{-1}): 1770, 1623, 1515, 1450, 1375, 1300, 1025, 830. NMR ($CDCl_3$, τ): 1·90 (*s*, H-2), 2·01 (*s*, H-5), 2·49 (*d*, *J* 8·5 Hz, H-2′, H-6′), 2·57 (*s*, H-8), 3·02 (*d*, *J* 8·5 Hz, H-3′, H-5′), 6·18 (*s*, OC$\underline{H}_3$), 7·69 (*s*, two OCOC$\underline{H}_3$).

6,7-*Dimethoxycoumarin*. M.p. 143–145° [lit.[20] m.p. 143–143·5°]. M found and required: 206. UV as required by lit.[20]

7-*Hydroxy*-4-*methoxy*-5-*methylcoumarin* (IIa). Needles, m.p. 297–300°. M found: 206·0604. $C_{11}H_{10}O_4$ requires: 206·0579. ν_{max}^{KBr} (cm^{-1}): 3150, 1700, 1620, 1565, 1500, 1270, 1160, 965, 825. λ_{max}^{EtOH} (nm): 225, 290 inf., 309, 320 inf. (ε 18 000, 11 300, 16 900, 15 000); $\lambda_{max}^{EtOH+NaOH}$ (nm): 238, 348 (ε 19 500, 23 600); $\lambda_{max}^{EtOH+NaOAc}$ (nm): 235, 322, 350 (ε 16 900, 13 000, 12 300). MS: M 206 (100%), *m/e* (%) 178 (49), 163 (46), 148 (14), 135 (18). *Methyl ether* (IIb). Obtained either with CH_2N_2–Et_2O or with Me_2SO_4–K_2CO_3–acetone as needles, m.p. and m.m.p. with a synthetic sample 194–196° (EtOH). ν_{max}^{KBr} (cm^{-1}): 3070, 1725, 1620, 1570, 1470, 1385, 1360, 1270, 1195, 1160, 1055, 980, 965, 850, 835, 825. RMN ($CDCl_3$, τ): 3·40 (*s*, H-6, H-8), 4·50 (*s*, H-3), 6·10, (*s*, OC$\underline{H}_3$), 6·20 (*s*, OC$\underline{H}_3$), 7·42 (*s*, CC$\underline{H}_3$). MS: M 220 (100%), *m/e* (%) 192 (62), 177 (44), 162 (11), 149 (17). *Acetate* (IIc). M.p. 187–189° (EtOH). ν_{max}^{KBr} (cm^{-1}): 3090, 1750, 1725, 1620, 1465, 1380, 1270, 1225, 1205, 1145, 1075, 1025, 970, 845. NMR ($CDCl_3$, τ): 3·08 and 3·25 (doublets, *J* 2·5 Hz, H-6, H-8), 4·40 (*s*, H-3), 6·08 (*s*, OC$\underline{H}_3$), 7·36 (*s*, CC$\underline{H}_3$), 7·71 (*s* OCOC$\underline{H}_3$). MS: M 248 (22%), *m/e* (%) 205 (100), 178 (44), 163 (15).

[27] R. M. HOROWITZ and L. JURD, *J. Org. Chem.* **26**, 2446 (1961).

Alkaline hydrolysis to 2,4-*dihydroxy*-6-*methylacetophenone*. A solution of IIa (25 mg) in 10% aq. KOH (5 ml) and dioxane (5 ml) was heated under reflux (2 hr), cooled to 0° and acidified with dil. HCl. The mixture was extracted with $CHCl_3$ (3 × 10 ml). The $CHCl_3$ soln was dried and evaporated, leaving an oily residue, which was chromatographed through a silica column, using $CHCl_3$ as eluant. The first fraction was recrystallized from $CHCl_3$, giving 2,4-*dihydroxy*-6-*methylacetophenone* (11 mg), m.p. and m.m.p. with an authentic sample 157–159° [lit.[23] m.p. 159°]. ν_{max}^{KBr} (cm^{-1}): 3175, 1620, 1575, 1465, 1275, 1170, 1000, 855, 825. *Alkaline hydrolysis to* 2,4-*dimethoxy*-6-*methylacetophenone*. A mixture of IIa (150 mg), 5% aq. NaOH (12 ml) and Me_2SO_4 (2 ml) was heated under reflux (24 hr). The mixture was cooled and extracted with $CHCl_3$ (3 × 10 ml). The $CHCl_3$-soln was dried and evaporated, giving 2,4-*dimethoxy*-6-*methylacetophenone* (69 mg), identified by direct comparison with an authentic sample [lit.[24] m.p. 48°]. ν_{max}^{KBr} (cm^{-1}): 2945, 2840, 1690 1602, 1580, 1470, 1320, 1255, 1200, 1160, 1100, 825. NMR ($CDCl_3$, τ): 3·73 (*s*, H-3, H-5), 6·25 (*s*, two $OC\underline{H}_3$), 7·59 (*s*, $CC\underline{H}_3$), 7·78 (*s*, $COC\underline{H}_3$). *Acid hydrolysis to* 4-*hydroxy*-7-*methoxy*-5-*methylcoumarin* (IId). The procedure was described by Desai and Sethna.[25] A suspension of IIb (25 mg) in conc. HCl–H_2O(1:1) (2 ml) was heated on the steam bath (0·5 hr). The mixture was cooled. The ppt. separated by filtration and recrystallized from EtOH, gave IId (8 mg), m.p. and m.m.p. with a synthetic sample 246–249°. ν_{max}^{KBr} (cm^{-1}): 3200–2500, 1705, 1615, 1355, 1320, 1270, 1200, 1160, 1040, 825. λ_{max}^{EtOH} (nm): 223, 290 inf., 309, 322 (ε 11 700 7200, 9400, 8000); $\lambda_{max}^{EtOH+NaOH}$ (nm): 227, 242, 300 (ε 13 000, 9700, 9000); $\lambda_{max}^{EtOH+NaOAc}$ (nm): 233, 300 (ε 7500 9000). *Synthesis of* 4-*hydroxy*-7-*methoxy*-5-*methylcoumarin* (IId). The procedure was described by Boyd and Robertson.[26] A mixture of 2-hydroxy-4-methoxy-6-methylacetophenone (120 mg),[23] diethyl carbonate (4 ml) and finely divided sodium (200 mg) was heated on the steam bath (20 min). After cooling, MeOH was added dropwise until excess sodium was destroyed, followed by Et_2O (20 ml). The mixture was extracted with H_2O (15 ml) and the soln acidified with HCl. The ppt. was separated by filtration and crystallized from EtOH giving IId (78 mg), m.p. 245–247°. *Methyl ether* (IIb). M.p. 194–196°.

7-O-*Glucosyloxy*-4-*methoxy*-5-*methylcoumarin* (IIe). M.p. 246–248°. ν_{max}^{KBr} (cm^{-1}): 3350, 1700, 1615, 1560, 1465, 1385, 1270, 1175, 815. λ_{max}^{EtOH} (nm): 225, 289, 306, 318 inf. (ε 23 500, 16 700, 18 400, 15 100); $\lambda_{max}^{EtOH+NaOH}$ (nm): 232, 289, 306, 318 inf. (ε 15 100, 15 100, 18 400, 14 000). No shift upon addition of NaOAc. MS: *m/e* (%) 206 (100), 178 (32), 163 (17), 148 (6), 135 (7). *Tetraacetate* (IIf). M.p. 176–178° (EtOH). ν_{max}^{KBr} (cm^{-1}): 1750, 1735, 1610, 1380, 1265, 1235, 1175, 1055, 850. NMR ($CDCl_3$, τ): 3·23 and 3·38 (doublets, *J* 2·5 Hz, H-6, H-8), 4·45 (*s*, H-3), 4·80 (*m*, glucose H-1, H-2, H-3, H-4), 5·80 (*m*, glucose $C\underline{H}_2$), 6·10 (*s*, $OC\underline{H}_3$, glucose H-5), 7·40 (*s*, $CC\underline{H}_3$), 7·90, 7·96 and 8·01 (singlets, 4 $OCOC\underline{H}_3$). MS: M 536 (< 1%), *m/e* (%) 331 (32), 207, (17), 206 (13), 169 (100), 127 (17), 109 (56). *Alkaline hydrolysis to* 7-O-*glucosyl*-4-*hydroxy*-5-*methylcoumarin* (IIg). A solution of IIe (150 mg) in aq. 10% NaOH (2 ml) was left at room temp. (0·5 hr) and acidified with conc. HCl. The precipitate was separated by filtration and crystallized from EtOH, giving IIg (106 mg), m.p. 185–188°. ν_{max}^{KBr} (cm^{-1}): 3350, 1665, 1610, 1560, 1330, 1180, 1085, 1055, 850, 825. λ_{max}^{EtOH} (nm): 219, 291, 308, 320 inf. (ε 16 800, 10 300, 11 500, 9200); $\lambda_{max}^{EtOH+NaOH}$ (nm): 235, 298 (ε 11 100, 10 300); $\lambda_{max}^{EtOH+NaOAc}$ (nm): 224, 300 (ε 13 700, 11 000). MS: *m/e* (%) 192 (100), 177 (9), 164 (11), 150 (92), 145 (16). *Pentaacetate* (IIh). Obtained from IIg as needles, m.p. 194–197° (EtOH). ν_{max}^{KBr} (cm^{-1}): 1780, 1755, 1735, 1610, 1370, 1260, 1240, 1170, 1070, 905. NMR ($CDCl_3$, τ): 3·18 and 3·30 (*d*, *J* 2·5 Hz, H-6, H-8), 3·85 (*s*, H-3), 4·78 (*m*, glucose H-1, H-2, H-3, H-4), 5·80 (*m*, glucose $C\underline{H}_2$), 6·10 (*m*, glucose H-5), 7·42 (*s*, $CC\underline{H}_3$), 7·64 (*s*, $OCOC\underline{H}_3$ at C-4), 7·90 (*s*, $OCOC\underline{H}_3$), 7·96 (*s*, two $OCOC\underline{H}_3$) and 7·99 (*s*, $OCOC\underline{H}_3$). *Acid hydrolysis to* 7-*hydroxy*-4-*methoxy*-5-*methylcoumarin* (IIa). A suspension of IIe (80 mg) in MeOH–conc. aq. HCl (9:1) (7 ml) was heated under reflux (4 hr). The mixture was cooled, the ppt. separated by filtration and identified with IIa by direct comparison. In the filtrate, glucose was identified by PC.

7-*Hydroxy*-4,8-*dimethoxy*-5-*methylcoumarin* (IIIa). M.p. 206–208° (toluene–AcOH, 1:1). M found: 236, $C_{12}H_{12}O_5$ requires 236. ν_{max}^{KBr} (cm^{-1}): 3300–2800, 1690, 1455, 1120, 805. λ_{max}^{EtOH} (nm): 223, 308 (ε 17 700 9500); $\lambda_{max}^{EtOH+NaOH}$ (nm): 225, 242, 266, 352 (ε 15 100, 16 400, 7100, 18 300). *Methyl ether* (IIIb). M.p. 180–182° (EtOH). ν_{max}^{KBr} (cm^{-1}): 1725, 1602, 1570, 1340, 1270, 1140, 1050. MS: M 250 (100%), *m/e* (%) 232 (33), 219, (28). *Acetate* (IIIc). M.p. 198–200° (EtOH). ν_{max}^{KBr} (cm^{-1}): 1752, 1730, 1600, 1580, 1420, 1220, 1025, 850. NMR ($CDCl_3$, τ): 3·31 (*s*, H-6), 4·43 (*s*, H-3), 6·10 (*s*, two $OC\underline{H}_3$), 7·48 (*s*, $CC\underline{H}_3$), 7·32 (*s*, $OCOC\underline{H}_3$).

Acknowledgements—This work was supported by the Conselho Nacional de Pesquisas, Brasil. The authors are indebted to Professor W. D. Ollis, The University, Sheffield, for the MS and to Dr. K. Zinner, University of São Paulo, for the ORD curves.