Phytochemistry, Vol. 28, No. 1, pp. 293–294, 1989.
Printed in Great Britain.

0031-9422/89 $3.00 + 0.00

COUMARINS OF THE FLOWERS OF *MURRAYA PANICULATA*

TIAN-SHUNG WU, MEEI-JEN LIOU and CHANG-SHENG KUOH*

Department of Applied Chemistry, Providence College of Arts and Sciences, Shalu 43309, Taichung Hsien, Taiwan, R.O.C.; *Department of Biology, National Cheng-Kung University, Tainan 70101, Taiwan, R.O.C.

(*Received* 12 *April* 1988)

Key Word Index—*Murraya paniculata* var. *omphalocarpa*; Rutaceae; coumarins; omphalocarpin; (−)-murracarpin; murrayacarpin-A and -B.

Abstract—Four new coumarins, omphalocarpin, (−)-murracarpin, murrayacarpin-A and -B together with known coumarins, scopolin, scopoletin, 5,7-dimethoxy-8-(3′-methyl-2′-oxobutyl)coumarin, (±)-murracarpin and mupanidin have been isolated from the flowers of *Murraya paniculata* var. *omphalocarpa*. Their structures have been characterized from spectral analysis, chemical transformation and/or synthesis.

INTRODUCTION

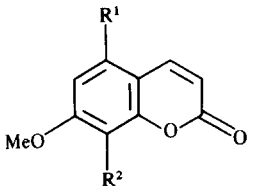
Murraya paniculata var. *omphalocarpa* Hayata (Rutaceae) has recently proved to be a rewarding source of a number of interesting new coumarins and flavonoids [1–3]. We were thus induced to examine the flower of this plant. The present work deals with the isolation and characterization of four new coumarins (**1**, **3**, **4**, and **5**) along with five known coumarins.

RESULTS AND DISCUSSION

Omphalocarpin (**1**) has a molecular formula $C_{17}H_{22}O_6$ based on the elemental analysis and mass spectrum [M^+, 322]. It showed UV absorption characteristics of a 7-alkoxycoumarin chromophore [4, 5]. It formed a acetyl derivative when treated with Ac_2O and pyridine, coupled with IR absorption at 3350, 1700 cm^{-1}, ^1H NMR spectrum at δ2.23 (disappeared on D_2O) indicating the presence of coumarin C=O and hydroxyl group in molecule. The ^1H NMR spectrum of **1** confirmed the presence of the 5,7-dimethoxy-8-substituted coumarin system [6, 7], showing the characteristic coumarin doublets at δ 6.14 (H-3) and 8.03 (1H, J = 9.4 Hz), a singlet at δ 6.34 (H-6) and a two aromatic methoxy singlet signal at δ 3.93 (C-5 and C-7). The remaining signals due to the C_5 unit side chain at C-8 comprise a doublet signal for benzylic methylene protons at δ2.94 (J = 6.0 Hz), an aliphatic methine proton at δ3.73 (q, J = 6.0 Hz, H-2′) and an aliphatic methoxy group at δ3.29 together with two methyl signal as a sharp singlet at δ1.28. Downfield shift of the methine proton at C-2′ to δ5.23 in omphalocarpin acetate (**2**) and mass fragmentation ions of **1** at m/z 205 $[M-C_6H_{13}O_2]^+$, 250 $[M-C_4H_9O+H]^+$ and 249 $[M-C_4H_9O]^+$, suggested the location of hydroxyl group at C-2′ and methoxy group at C-3′. The side chain was thus identified as 2′-hydroxy-3′-methoxy-3′-methylbutyl attached to C-8 through C-1′ leading to the expression **1** for omphalocarpin.

(−)-Murracarpin (**3**) mp 164–165°, $[\alpha]_D$ −15.6° ($CHCl_3$), $C_{16}H_{18}O_5$, showed UV absorption maxima at 210.5, 248.8, 258.3 and 322.6 nm characteristic of a 7-oxygenated-8-substituted coumarin moiety [4, 5]. The IR spectrum exhibited strong absorption at 3510 (OH), 1730 (lactonic carbonyl), 1615, 1570 and 1510 cm^{-1} (aromatic nucleus). The presence of alcoholic hydroxyl group and the absence of a phenolic hydroxyl function was indicated by the negative ferric chloride reaction. The ^1H NMR spectrum of **3** displayed characteristic signals for a methyl group (δ1.87, *s*), an alcoholic hydroxyl function (δ2.55, *br*, exchangeable with D_2O), a benzylic methine proton (δ5.19, *d*, J = 6.7 Hz), an aliphatic methine signal (δ4.80, *br d*, J = 6.7 Hz), an exo-methylene group (δ4.87 and 4.97, each 1H, *m*), two aromatic *ortho* located protons (δ7.42 and 6.90, each 1H, *d*, J = 8.7 Hz), and the C-3 and C-4 protons of the coumarin nucleus (δ6.26 and 7.63, each 1H, *d*, J = 9.5 Hz) an aromatic methoxy group (δ3.95), and an aliphatic methoxy signal (δ3.30). The location of aliphatic methoxy group at C-1′ and hydroxyl group at C-2′ was suggested from the mass fragmentation ions at m/z 273 $[M-OH]^+$ and 219 $[M-C_4H_7O]^+$. Thus murracarpin has structure **3**. This is the first report of the occurrence of **3** as an optically active compound in a natural source. However, the racemate of **3*** has been isolated from *Murraya exotica* [8], *Phebalium tuberculosum* [9] and has been synthesized [10]. Murrayacarpin-A (**4**) was isolated as colourless needles, mp 163–166°. The molecular formula was established as $C_{11}H_{10}O_4$ by HRMS. The UV spectrum of **4** was very similar to that of **3** characteristic of a typical 7-oxygenated-8-substituted coumarin nucleus [4, 5]. The IR spectrum of **4** showed

*We have assigned a trivial name for convenience.



1 R^1 = OMe, R^2 = $CH_2CH(OH)C(Me)_2OMe$
2 R^1 = OMe, R^2 = $CH_2CH(OAc)C(Me)_2OMe$
3 R^1 = H, R^2 = $CH(OMe)CH(OH)C(Me)=CH_2$
4 R^1 = H, R^2 = CH_2OH
5 R^1 = OMe, R^2 = CH_2OH
6 R^1 = OMe, R^2 = CHO

absorption peaks at 3650 (OH), 1710 (lactonic C=O), and 1650 (aromatic C=C). The ^{1}H NMR spectrum of murrayacarpin-A appeared signals for H-3 and H-4 at δ6.27 and 7.64 (each 1H, *d*, J = 9.5 Hz), two mutually *ortho* coupling aromatic protons at δ6.88 and 7.42 (each 1H, *d*, J = 8.5 Hz), a methoxy group at δ 3.97, signals at δ4.97 (2H, *d*, J = 6.7 Hz) and 2.41 (1H, *t*, J = 6.7 Hz, exchangeable with D_2O) due to a hydroxymethyl group. According to the above data, murrayacarpin-A is **4**.

Murrayacarpin-B (**5**), mp 199–201°, $C_{12}H_{12}O_5$. Its UV spectrum was characteristic of a 5,7-dioxygenated-8-substituted coumarin [4, 5]. The ^{1}H NMR spectrum of **5** was very similar to that of **4** except a methoxy group at δ3.95 and a singlet signal at δ 6.33 (H-6) instead of a pair doublet signals at δ 6.88 and 7.42 in **4**. The mass spectrum of **5** showed a similar breakdown pattern with **4** and appeared a parent ion at *m/z* 236. Thus, murrayacarpin-B can be represented by structure **5**. Further evidence in support of this structure has been adduced by the reduction of 8-formyllimettin (**6**) which was prepared from the formylation of limettin (**7**) with $POCl_3$ and DMF.

Known coumarins, scopolin (**8**) [11], scopoletin (**9**) [11], 5,7-dimethoxy-8-(3′-methyl-2′-oxobutyl)coumarin (**10**) [2], (±)-murracarpin (**11**) [3,8–10] and mupanidin (**12**) [3, 12, 13] were also isolated and identified by the spectral comparison and/or mmp with authentic samples.

EXPERIMENTAL

Mps: uncorr.; ^{1}H NMR: $CDCl_3$ except where noted, TMS as int. stand; MS: direct inlet; UV: MeOH; IR: KBr, unless otherwise stated.

Plant material. Murraya paniculate var. *omphalocarpa* Hayata was collected from Orchid Island (Lan-Yu) in Sept. 1985 and verified by Prof. C.-S. Kuoh. A specimen is deposited in the Herbarium of Cheng-Kung University, Taiwan, Republic of China.

Extraction and separation. The fresh flowers (260 g) was extracted with MeOH (×3) and evapd to dryness. The MeOH extract was dissolved in Me_2CO to yield **8** (6 g). The filtrate after remove of **8** was adsorbed on silica gel, transfered to a silica gel column packed in C_6H_6–Me_2CO (9:1) and eluted with a gradient of C_6H_6 and Me_2CO to afford 7 fractions. The fraction 1 was rechromatographed on a silica gel column with *n*-hexane–EtOAc (5:1) to give **10** (10 mg) and **a** (2 mg), respectively. A colourless crystal **9** (50 mg) was obtained from fraction 3. Fraction 5 was repeatedly chromatographed on 10% $AgNO_3$ silica gel with C_6H_6–Me_2CO (9:1) as eluant to yield **1** (15 mg), **11** (30 mg) and **3** (4 mg), successively. The fraction 7 was separated by prep. TLC. (silica gel) using a solution of $CHCl_3$–Me_2CO (20:1) and then purified by 10% $AgNO_3$ silica gel prep. TLC (EtOAc as developing solvent) to afford **4** (1 mg), **5** (2 mg) and **12** (6.5 mg), respectively.

Omphalocarpin (**1**). Colourless needles ($CHCl_3$–Me_2CO), mp 159–160°. Calcd for $C_{17}H_{22}O_6$: C, 63.34; H, 6.88. Found: C, 63.38; H, 6.83; $[\alpha]_D$ −44.1° (c, 0.3, $CHCl_3$). UV λ_{max} nm (log ε): 238.2 (3.79), 260.9 (4.01), 326.7 (4.4), IR ν_{max} cm^{-1}: 3350, 1700, 1600, 1500; MS *m/z* (rel. int.): 322 (M^+, 11), 304 (8), 250 (40), 249 (28), 220 (34), 219 (75), 207 (100), 205 (32), 189 (16), 177 (11), 161 (45), 73 (54). ^{13}CNMR: δ 20.2 (*q*), 20.9 (*q*), 24.5 (*t*), 49.2 (*q*), 55.8 (*q*), 56.0 (*q*), 56.0 (*q*), 77.1 (*d*), 78.3 (*s*), 90.3 (*d*), 103.7 (*s*), 107.9 (*s*), 110.6 (*d*), 130.7 (*d*), 154.0 (*s*), 155.5 (*s*), 161.2 (*s*). Acetate, colourless granules **2**, mp 111–113° ($CHCl_3$); UV λ_{max} nm: 208.5, 250.5, 259.8 and 325.6; MS *m/z*: 364 (M^+), 304, 289, 273, 249, 232, 219, 205, 189, 161, 73 (100%); ^{1}H NMR: δ 1.25 (3H, *s*, Me), 1.28 (3H, *s*, Me), 1.80 (3H, *s*, OAc), 2.96 (1H, *dd*, J = 3.3 and 14 Hz, H-1′), 3.16 (1H, *dd*, J = 9.3 and 14 Hz, H-1′), 3.33 (3H, *s*, 3′-OMe), 3.91 (3H, *s*, OMe), 3.93 (3H, *s*, OMe), 5.23 (1H, *dd*, J = 3.3 and 9.3 Hz, H-2′), 6.12 (1H, *d*, J = 9.6 Hz, H-3), 6.28 (1H, *s*, H-6), 7.97 (1H, *d*, J = 9.6 Hz, H-4).

(−)-*Murracarpin* (**3**). Colourless granules (*n*-hexane), mp 164–165°, $C_{16}H_{18}O_5$; $[\alpha]_D$ −15.6° ($CHCl_3$; *c* 0.068); UV λ_{max} nm (log ε): 210.5 (4.63), 248.8 (4.00), 258.3 (4.01), 322.6 (4.53); MS *m/z* (rel. int.): 273 (M^+ − OH, 1), 258 (M^+ − MeOH, 1), 220 (14), 219 (100), 205 (9), 189 (9), 175 (7), 161 (17), 131 (6).

Murrayacarpin-A (**4**). Colourless needles ($CHCl_3$), mp 163–166° (dec.). Calcd for $C_{11}H_{10}O_4$ [M]$^+$ = *m/z* 206.0578; Found: 206.0548. UV λ_{max} nm (log ε): 246.4 (3.48), 256.8 (3.47), 321.9 (4.06); MS *m/z* (rel. int.): 206 (M^+, 100), 205 (31), 177 (53), 175 (28), 174 (61), 163 (22), 146 (25), 131 (25).

Murrayacarpin-B (**5**). Colourless granules ($CHCl_3$), mp 199–201°. Calcd for $C_{12}H_{12}O_5$ [M]$^+$ = *m/z* 236.0684; Found: 236.0746. UV λ_{max} nm (log ε): 208.4 (4.31), 250.5 (3.62), 259.0 (3.66), 326.2 (3.85); MS *m/z* (rel. int.): 236 (M^+, 100), 235 (66), 220 (23), 219 (64), 207 (45), 205 (23), 204 (32), 161 (36).

Synthesis of murrayacarpin-B (**5**). Freshly distilled $POCl_3$ (2 ml) wass added to limettin (**7**) (1.29 g) in 5 ml DMF. The mixture was refluxed at 80° for 8 hr. The reaction mixture was poured into ice 5% K_2CO_3 solution and extracted with $CHCl_3$. The $CHCl_3$ layer was dried and evapd to afford a yellow residue which was recrystallized with MeOH to give 8-formylimettin (**6**) as yellow crystals (0.64 g), mp 170° (sublim.). UV λ_{max} nm: 216.6, 282.4, 321.4; MS *m/z*: 234 (M^+, 100%), 206, 181, 177, 163, 133. ^{1}H NMR: δ 4.02 (6H, *s*, 2× OMe), 6.24 (1H, *d*, J = 9.8 Hz, H-3), 6.32 (1H, *s*, H-6), 7.97 (1H, *d*, J = 9.8 Hz, H-4), 10.54 (1H, *s*, 8-CHO). **6** (100 mg) in MeOH was reduced by excess $NaBH_4$ at room temp. overnight. The reaction mixture was worked-up in the usual way to give colourless crystals (30.4 mg), mp 222–223° ($CHCl_3$); calcd. for $C_{12}H_{12}O_5$: C, 61.01; H, 5.12. Found: C, 60.73; H, 5.13%. This compound was identified with the natural **5** by comparison of ^{1}H NMR, IR, MS spectra and TLC.

Acknowledgements—We thank Dr M. Niwa (Meijo University, Japan) for measurement of the high resolution mass spectrum. This investigation was supported by a grant (NSC76-0208-M126-06) from the National Science Council of the Republic of China.

REFERENCES

1. Wu, T. S. (1981) *Phytochemistry* **20**, 178.
2. Wu, T. S., Tien, H. J., Arisawa, M., Shimizu, M. and Morita, N. (1980). *Phytochemistry*, **19**, 2227.
3. Wu, T. S. (1988) *Phytochemistry* **27**, 2357.
4. Stanly, W. L., Waiss, A. C. Jr., Lundin, R. E. and Vannier, S. H. (1965) *Tetrahedron* **21**, 89.
5. Smith, E., Hosansky, N., Bywater, W. G. and Van Tamelen, E. E. (1957), *J. Am. Chem. Soc.*, **79**, 3534.
6. Fisher, J. F. and Nordby, H. E. (1966) *Tetrahedron* **22**, 1489.
7. Steck, W. and Mazurek, M. (1972) *Lloydia* **35**, 418.
8. Manandhar, M. D. (1980). *Indian J. Chem.* **19B**, 1006.
9. Chow, P. W., Duffield, A. M. and Jefferies, P. R. (1966) *Aust. J. Chem.* **19**, 483.
10. Imai, F., Kinoshita, T., Itai, A. and Sankawa, U. (1986) *Chem. Pharm. Bull.* **34**, 3978.
11. Wu, T. S., Lin, C. N., Yang, L. K. and Lin, S. T. (1975) *J. Chin. Chem. Soc.* **22**, 163.
12. Talapatra, S. K., Dutta, L. N. and Talapatra, B. (1973) *Tetrahedron* **29**, 2811.
13. Kong, Y. C., Cheng, K. F., Ng, K. H., But, P. P.-H., Li, Q., Yu, S. X., Chang, H. T., Cambie, R. C., Kinoshita, T., Kan, W. S. and Waterman, P. G. (1986) *Biochem. Syst. Ecol.* **14**, 491.