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7-METHOXYCOUMARINS FROM *MICROMELUM MINUTUM*

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Key Word Index—*Micromelum minutum*, Rutaceae, 7-methoxycoumarins, osthol, micromelin, murralongin, murrangatin, dihydromicromelins A and B, acetyldihydromicromelin A, minumicrolin, 7,12-ether of 5,7-dihydroxy-3,6,8,4′-tetramethoxyflavone, murrangatin

Abstract—Chemical investigation of an Assamese collection of *Micromelum minutum* gave four known coumarins (osthol, micromelin, murralongin and murrangatin) and five new coumarins, dihydromicromelin A and B, acetyldihydromicromelin A, the *threo* diastereomer of murrangatin, the 7,12-ether of 5,7-dihydroxy-3,6,8,4′-tetramethoxyflavone and murrangatin

INTRODUCTION

Previous chemical studies of *Micromelum* species (Rutaceae) have revealed the presence of 6- or 8-prenylated 7-methoxycoumarins such as micromelin (micromelumin, **1**) or microminutin (**2**) in which the side chain is modified in unusual ways [1–5, 7, 8] Some simpler coumarins and a few alkaloids typical of Rutaceae have also been found [6–8] We now describe the isolation from Assamese *M minutum* (Forst f) Wight and Arn of **1**, osthol (**3**), murralongin (**4**) [9], murrangatin (**5a**) [10–12] and the following new coumarins—the hemiacetals dihydromelin A and B as a mixture of C-5′ epimers **6a** and **6b**, the acetate **6c** derived from dihydromicromelin A, minumicrolin (**7a**) which is a diastereomer of murrangatin, and the unusual ether **8a*** Isolation of **1** and **3** from a Northern Queensland collection [1] and isolation of **2** and flindersin from a Thai collection of *M minutum* [8] have been reported earlier†

RESULTS AND DISCUSSION

The ^{1}H NMR spectrum (see Experimental) of acetyldihydromicromelin A (**6c**), $C_{17}H_{16}O_7$ (high resolution mass spectrum), mp 66–68°, showed that it was a 6-substituted 7-methoxycoumarin The nature of the oxidized prenyl side chain was at first somewhat obscure as H-11 at δ5 43 and H-12 at δ3 83 were singlets and not coupled to each other, until it was realized that a similar situation prevails in micromelin (**1**) As the spectrum of the new substance also exhibited methyl singlets at 2 17 (acetate) and 1 51 (MeC–O) and a low field one proton singlet at δ6 38, the structure was expanded to **6c** This was confirmed by hydrolysis (K_2CO_3–MeOH–H_2O) to a mixture of **6a** and **6b** whose spectra differed from each other principally in the chemical shifts of H-5 (δ7 46 in **6a** vs δ7 82 in **6b**) and H-14 (δ5 41 *br* in **6a** vs δ5 52 *br* in **6b**) These two substances also occurred in the plant, reacetylation of the mixture afforded only **6c**, whereas oxidation gave micromelin The C-14 stereochemistry assigned to **6a** and **6c**, on the one hand, and **6b**, on the other, is based on the significant paramagnetic shift of the H-5 signal of **6b** ($\Delta\delta$0 36) compared with H-5 in **6a** and **6c** which necessitates β-orientation of the hydroxyl group in **6b** (model)

Minumicrolin (**7a**), $C_{15}H_{16}O_5$, mp 132–135°, $[\alpha]_D$ + 17 5°, was deceptively similar to murrangatin (**5a**) mp 132–133°, $[\alpha]_D$ − 17°, which has been previously found in *Murraya elongata* [10] and *Murraya paniculata* [11, 12] and was the major coumarin constituent in our collection of *M minutum* However, that it was not merely the enantiomer of **5a** was evident from close inspection of the ^{1}H NMR spectra of **5a** and **7a** (see Experimental) which differed significantly in the chemical shifts of the vinylic

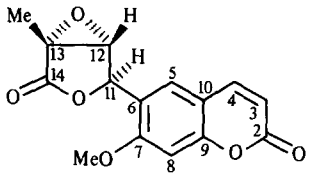
Table 1 ^{13}C NMR spectrum of **6c** ($CDCl_3$, 67 89 MHz)*

C-2	159 30	C-11	77 93 *d*
3	113 73 *d*	12	63 63 *d*
4	143 20 *d*	13	64 44
5	126 76 *d*	14	96 89 *d*†
6	123 34	15	12 50 *q*
7	160 81	OMe	56 17 *q*
8	98 92 *d*	OAc	169 30
9	155 85		21 23 *q*
10	112 03		

*Unmarked signals are singlets
†Assignment by selective decoupling

*The absolute configurations of **1**, **5a**, **6a**, **7a** and **8a** are unknown

†Refs [1] and [8] refer to this taxon as *M minutum* (Forst f) Seem, presumably because it is so listed in several floras (see also Englert, A and Prantl, *Die Naturlichen Pflanzenfamilien*, 2nd edn, Leipzig, Wilhelm Engelmann Verlag, (1931) Vol 19a, p 318) We think this is based on misinterpretation of an entry in *Index Kewensis*, (1895) Vol II, p 231, Clarendon Press, Oxford, see also Supplementary Volume XIV, p 87 (1970)



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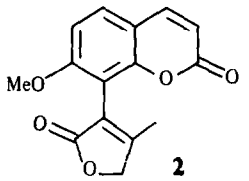

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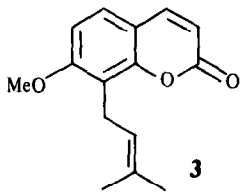

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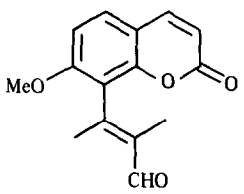

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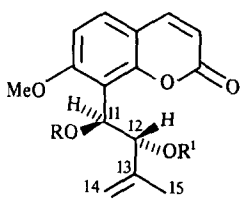
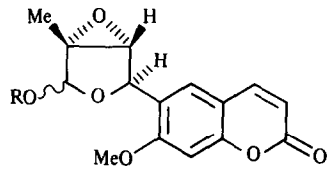
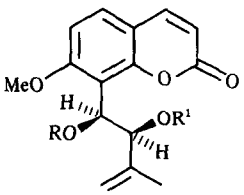


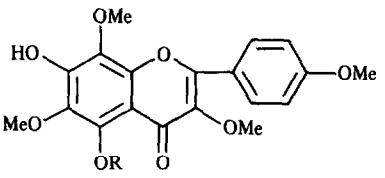
5a R, R^1 = H
5b R, R^1 = Ac
5c R = C_6H_5CO, R^1 = H
5d R, R^1 = C_6H_5CO
5e R = H, R^1 = Et
5f R = Ac, R^1 = Et



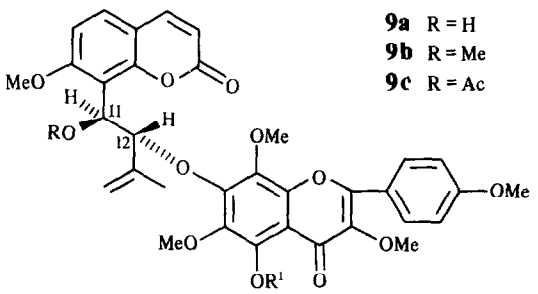
6a R = α-OH
6b R = β-OH
6c R = α-OAc



7a R, R^1 = H
7b R, R^1 = Ac
7c R, R^1 = C_6H_5CO



9a R = H
9b R = Me
9c R = Ac



8a R, R^1 = H
8b R = H, R^1 = Me
8c R, R^1 = Ac

protons in the prenyl side chain, those of **7a** appearing 0 4 ppm downfield from those of **5a** The same relationship was found in the NMR spectra of the diacetates **5b** (mp 123–124°) and **7b** (gum) and the dibenzoates **5c** (mp 100–102°) and **7c** (mp 225–227°) As the *erythro* configuration **5a** has been deduced for murrangatin [10], minumicrolin must also be the *threo* isomer **7a**

That **8a**, $C_{34}H_{34}O_{12}$ (high resolution mass spectrum), mp 182–186°, had an oxygen bridge linking C-12 of murrangatin and C-7 of 5,7-dihydroxy-3,6,8,4'-tetramethoxyflavone was deduced as follows The ^{1}H NMR spectrum (Table 2) was a composite of the NMR spectra of murrangatin, with the signals of H-11 and H-12 displaced toward lower field, and of a 5-hydroxy-3,6,7,8,4'-pentaalkoxyflavone containing four methoxyl groups* Methylation of **8a** to **8b** (CH_2N_2–MeOH) resulted in the disappearance of the 5-hydroxyl signal,

*In the literature [13, 14 and numerous references cited therein] the signals of protons at C-2', C-3', C-5' and C-6' of 4-oxygenated flavones and flavonols are generally described as two pairs of *ortho*-coupled doublets This is not correct as H-2' and H-3', or H-5' and H-6', are magnetically non-equivalent Indeed, examination of **8a–c** and **9a–c** at high resolution showed the complex pattern due to an AA'XX' system

Table 2 ^{1}H NMR spectra of **8a–8c** ($CDCl_3$, 270 MHz)*

	8a	8b	8c
H-3	6 26 (*d*, 10)	6 25 (*d*)	6 27 (*d*)
4	7 63 (*d*, 10)	7 62 (*d*)	7 61 (*d*)
5	7 40 (*d*, 8)	7 40 (*d*)	7 40 (*d*)
6	6 85 (*d*, 8)	6 83 (*d*)	6 81 (*d*)
11	5 77 (*d*, 9)	5 74 (*d*)	6 49 (*m*)
12	5 33 (*dbr*, 9)	5 37 (*dbr*)	6 49 (*m*)
14a	4 71 (*br*)	4 74 (*br*)	4 99 (*br*)
14b	4 64 (*quint*, 1 5)	4 63 (*quint*)	4 71 (*quint*)
15	1 69 (*br*)	1 67 (*br*)	1 57 (*br*)
2′,6′	8 12 (*dm*, 9)	8 80 (*dm*)	8 10 (*dm*)
3′,5′	7 03 (*dm*, 9)	7 01 (*dm*)	7 03 (*dm*)
OH	12 44, 4 51	4 45	—
OMe†	3 90, 3 90	3 90, 3 88	3 93, 3 89
	3 85, 3 85	3 88, 3 86	3 89, 3 85
	3 73	3 85, 3 75	3 78
OAc	—	—	2 46, 1 98

*Unmarked signals are singlets

†In C_6D_6 for **8b** 3 95, 3 80, 3 70, 3 70, 3 28 and 3 08, for **8c** 4 01, 3 85, 3 69, 3 31 and 3 28

while formation of diacetate **8c** was also accompanied by a paramagnetic shift of the H-11 signal Hence, the ether linkage involved the hydroxyl on C-12 of the coumarin moiety

The mass spectra of **8a–c** exhibited only weak peaks corresponding to $[M]^+$, the main feature being cleavage, with hydrogen transfer, to an ion of *m/z* 258 ($C_{15}H_{14}O_4^+$) which emanated from the murrangatin half and ions of *m/z* 374, 388 or 416 which corresponded to flavones **9a**, **9b** or **9c**, respectively Further fragmentation of the latter gave rise, in all cases, to a significant peak at *m/z* 135 (B_2), indicating that C-4′ of **8a** carried a methoxyl group [13] This was confirmed by the following observation In the NMR spectra of **8a–c**, large benzene-induced upfield shifts were observed for two methoxyl signals of which one could be attributed to the methoxyl at C-7 of the coumarin moiety The other could only be assigned to a methoxyl on C-4′ of the flavonol half since a C-3 methoxyl does not show a large benzene-induced solvent shift [13]

Hydrolysis of **8a** (KOH–EtOH–H_2O) gave a 2 1 mixture of coumarin **5e** and flavone **9a** by the path shown in Scheme 1 which involves a double inversion at C-12 of the coumarin half Separation of the hydrolysis mixture was accomplished by way of the acetates **5f** and **9c** The chemical shifts of the vinylic protons of **5e** and **5f** showed that these compounds, like **8a–c**, belonged to the murrangatin series The NMR spectra of **9a** and **9c** coincided with the spectra of authentic 5,7-dihydroxy-3,6,8,4′-tetramethoxyflavone and its diacetate The former has been isolated from *Ambrosia grayi* [15] Mass spectra of **9c** and the authentic diacetate were also identical Consequently the ether linkage of **8a** involved the hydroxyl on C-7 of the flavone moiety

EXPERIMENTAL

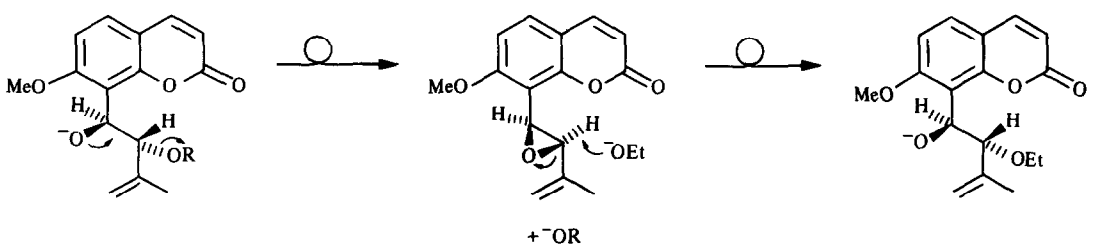
Isolation of constituents Above ground parts of *M minutum* (Forst f) Wight and Arn (1 5 kg) collected in the Diphu area of the Karbi Anglong District, Assam, India (voucher on deposit in herbarium of the RRL, Jorhat) were extracted with $CHCl_3$ in a Soxhlet apparatus until the extract was colorless After removal of solvent at red pres the residue (60 g) was dissolved in 400 ml of MeOH containing 10% H_2O, allowed to stand overnight and filtered The filtrate was washed with petrol (60–80°, 6 × 300 ml), the MeOH portion was concd at red pres and the residue thoroughly extracted with $CHCl_3$ (8 × 200 ml) Evapn of the washed and dried extract furnished 26 g of a gummy residue which was chromatographed over 550 g of silica gel (60–120 mesh, BDH), 200 ml fractions being collected as follows Frs 1–12 (C_6H_6), 13–52 (C_6H_6–EtOAc, 4 1), 53–103 (C_6H_6–EtOAc, 4 1), 104–142 (C_6H_6–EtOAc, 2 1), 143–191 (C_6H_6–EtOAc, 1 1), 192–204 (EtOAc), 205–216 (EtOAc–MeOH, 19 1), 217–224 (EtOAc–MeOH, 9 1), 225–238 (EtOAc–MeOH, 4 1), 239–247 (EtOAc–MeOH, 2 1)

Fr 19–26 (600 mg) were combined Purification by prep TLC (C_6H_6–EtOAc, 14 1, two developments) gave 80 mg of osthol (**3**) Fr 50–56 (400 mg) on purification by prep TLC (C_6H_6–EtOAc, 4 1, thickness of plates 0 75 mm) gave 60 mg of micromelin (**1**) mp 216–218° (Et_2O), lit [7] mp 218–219° (EtOH) IR and ^{1}H NMR data were in agreement with published values [1, 2, 5]

Fr 57–74 (0 7 g), which exhibited one major spot, were combined and purified by prep TLC (C_6H_6–EtOAc, 9 1, two developments) to yield 0 5 g of **6c**, mp 66–68° (EtOAc–C_6H_6), IR $\nu_{max}^{CHCl_3}$ cm^{-1} 1730 (br), 1625, 1275, 1130, 1085, 1065, 1000, 975, 940, 890, 850 and 825, ^{1}H NMR ($CDCl_3$) δ 6 29 (*d*, *J* = 10 Hz, H-3), 7 60 (*d*, *J* = 10 Hz, H-4), 7 50 (H-5), 6 84 (H-8), 5 43 (H-11), 3 83 (H-12), 6 38 (H-14), 1 51 (H-15), 3 96 (OMe) and 2 17 (Ac) [Calc for $C_{17}H_{16}O_7$ MW, 332 0895 Found MW (MS), 332 0886] Other significant ions in the HR-MS were at *m/z* (composition, %) 273 ($C_{15}H_{13}O_5$, 19), 229 ($C_{13}H_9O_4$, 100) and 213 ($C_{13}H_{19}O_3$, 71)

Fr 80–85 (2 7 g), which exhibited one major spot, were combined and purified by prep TLC (C_6H_6–EtOAc, 9 1, two developments) to give 0 4 g of murralongin (**4**) as a gum, MS *m/z* 258 ($[M]^+$), 243, 229, 215 and 199 IR and ^{1}H NMR data were in agreement with published values [9]

Fr 86–120 (1 63 g), which exhibited two major spots, were combined and purified by prep TLC (C_6H_6–EtOAc, 4 1, three



Scheme 1 Hydrolysis products of **8a**

developments) The faster moving band yielded 0 1 g of **8a** as a yellow solid, mp 182–186° (EtOAc), IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3500, 1725, 1600, 1290, 1160, 1120 and 1000, ^{1}H NMR Table 2, MS *m/z* (rel int) 632 (1 8) [M]$^+$, 374 (100), 373 (6 0), 359 (72 2), 341 (6 8), 258 (5 5), 231 (4 8), 229 (4 3), 213 (5 2), 203 (5 9), 199 (5 0), 197 (5 0), 18 g (28 2), 148 (6 7), 136 (16 9), 131 (13 4) [Calc for $C_{34}H_{32}O_{12}$ MW, 632 1890 Found MW (MS), 632 1879] A significant ion in the HR-MS corresponded to **9a** [Calc for $C_{19}H_{18}O_8$ MW, 374 0999 Found MW (MS), 374 0997] Methylation of 20 mg of **8a** in 2 ml of MeOH with CH_2N_2, destruction of excess CH_2N_2 after 1 hr by addition of a few drops of HOAc, removal of solvent at red pres followed by removal of HOAc by co-distillation with toluene and prep TLC of the residue (C_6H_6–EtOAc, 1 1) gave as the major product **8b** as a gum, IR ν_{max} cm^{-1} 3415, 3350, 1750, 1705, 1600, 1185, 1160, 1115, 1060, 950, ^{1}H NMR spectrum Table 2, MS *m/z* (rel int) 646 (0 2) [M]$^+$, 575 (2 1), 388 (86 6), 387 (25 0), 373 (100), 369 (10 1), 359 (10 7), 355 (42 4), 345 (3 3), 343 (4 1), 341 (4 4), 330 (13 4), 315 (6 1), 287 (3 4), 273 (2 3), 258 (10 0), 231 (8 3), 229 (7 0), 213 (8 9), 211 (7 4), 203 (9 0), 189 (52 6), 159 (11 1), 135 (24 2), 131 (22 6) [Calc for $C_{35}H_{34}O_{12}$ MW, 626 2047 Found MW (MS), 646 2013] A significant ion in the HR-MS corresponded to **9b** [Calc for $C_{20}H_{20}O_8$ MW, 388 1156 Found MW (MS), 388 1156] A minor, less polar fraction was identified by its ^{1}H NMR spectrum as 5-hydroxy-3,6,7,8,4′-pentamethoxyflavone (calycopterin 4′-methyl ether, **9b**), ^{1}H NMR ($CHCl_3$, 270 MHz) δ 12 60 (OH), 8 19 (*dm*, *J* = 9 Hz, H-2′, H-6′), 7 07 (*dm*, *J* = 9 Hz, H-3′, H-5′), 4 13, 3 99, 3 99, 3 93, 3 89 (OMe), (C_6D_6) 8 15 (*d*, H-2′, H-6′), 6 76 (*d*, H-3′, H-5′), 3 88, 3 84, 3 74, 3 69, 3 25 (OMe) It is not clear whether this substance was formed by hydrolysis of **8a** followed by methylation or by methylation of an impurity accompanying **8a**

Acetylation of 10 mg of **8a** with pyridine–Ac_2O, work-up in the usual fashion and purification by prep TLC (C_6H_6–EtOAc, 2 1) afforded 8 mg of the diacetate **8c** as a gum, IR $\nu_{max}^{CHCl_3}$ cm^{-1} 1730, 1605, 1175, 1115, 1025, ^{1}H NMR Table 2, MS *m/z* (rel int) 717 (0 04) [M + 1]$^+$, 416 (1 4), 374 (4 9), 359 (7 5), 345 (7 8), 301 (92 2), 259 (100), 241 (5 4), 231 (80 0), 203 (15 7), 189 (47 6), 135 (8 5), 131 (7 2)

The slower moving band from fr 86–120 yielded 80 mg of a mixture of **6a** and **6b** as a gum (EtOAc), IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3500, 1725, 1620, 1260, 1125, 1080, 960, 940, 900, MS *m/z* 290 [M]$^+$, 272, 255, 222, 192 The ^{1}H NMR spectrum of the mixture exhibited the same peaks as the mixture prepared by hydrolysis of **6c** (*vide infra*), but the proportion of the two components was somewhat different In one expt, further purification of the **6a, b** mixture by TLC afforded a solid, mp 160–163° (EtOAc), possibly one of the isomers, but the NMR spectrum of this material was not recorded

Fr 140–160 (3 78) were mixtures Purification by prep TLC (C_6H_6–EtOAc, 2 3, three developments) gave 0 30 g of the *threo*-isomer **7a** of murrangatin, mp 132–135° (EtOAc–MeOH), $[\alpha]_D$ + 17 50 ($CHCl_3$, 0 428 8/100 ml), IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3500, 1730, 1600, 1560, 1275, 1160, 1115, 985, 900, 825, ^{1}H NMR ($CDCl_3$, 270 MHz) δ 6 26 (*d*, *J* = 10 Hz, H-3), 7 64 (*d*, *J* = 10 Hz, H-4), 7 41 (*d*, *J* = 8 Hz, H-5), 6 90 (*d*, *J* = 8 Hz, H-6), 5 42 (*d*, *J* = 9 Hz, H-11), 4 53 (*dbr*, *J* = 9 Hz, H-12′), 4 99 (*br*, H-14a, b), 1 90 (*br*, H-15), 3 98 (OMe), MS *m/z* 276 [M]$^+$, 258, 205, 175 Acetylation of 15 mg of **7a** with pyridine–Ac_2O followed by the usual work-up and purification by TLC (C_6H_6–EtOAc, 4 1) gave 12 mg of diacetate **7b** as a gum, IR $\nu_{max}^{CHCl_3}$ cm^{-1} 1730, 1605, 1560, 1285, 1250, 1175, 1115, 1025, 900, ^{1}H NMR ($CDCl_3$, 270 MHz) δ 6 27 (*d*, H-3), 7 60 (*d*, H-4), 7 40 (*d*, H-5), 6 84 (*d*, H-6), 6 59 (*d*, H-11), 5 93 (*dbr*, H-12), 5 06 (*br*) and 4 99 (*quint*, *J* = 2 Hz, H-14a, b), 1 89 (*br*, H-15), 3 96 (OMe), 2 06, 1 82 (Ac) [Calc for $C_{19}H_{20}O_7$ MW, 360 1207 Found MW (MS), 360 1226] Other significant ions in the LR-MS were at *m/z* (%) 300 (7), 258 (9), 247 (10), 205 (100) and 175 (7) Benzoylation of 20 mg of **7a** (benzoyl chloride–pyridine) followed by usual work-up and purification by prep TLC (C_6H_6–EtOAc, 14 1, two developments) gave 25 mg of **7c**, mp 225–227° (MeOH–$CHCl_3$), IR ν_{max}^{KBr} cm^{-1} 1725 (br), 1600, 1535, 1265, 1175, 1100, 1065, 1025, 825, 700, ^{1}H NMR ($CDCl_3$, 270 MHz) δ 8 61 (*d*, *J* = 8 Hz) and 7 88 (*d*, *J* = 8 Hz, each two aromatic protons), 7 61–7 25 (complex *m* of H-4, H-5 and six aromatic protons), 6 27 (*d*, H-3), 6 77 (*d*, H-6), 7 01 (*d*, H-11), 6 22 (*d*, H-12), 5 33 (*br*) and 5 09 (*quint*, H-14a, b), 3 91 (OMe) and 1 99 (H-15), CD curve (MeOH) $[\theta]_{318}$ −4820 (min), $[\theta]_{311}$ −2800 (max), $[\theta]_{288}$ −4100 (min), $[\theta]_{270}$ 0 (max), $[\theta]_{247}$ −20700 (min), $[\theta]_{230}$ 0 (max), $[\theta]_{215}$ −44000 (last reading)

Fr 161–194 (3 66 g), which exhibited one major spot, were combined Purification by prep TLC ($CHCl_3$–MeOH, 15 1) gave 2 5 g of murrangatin (**5a**), mp 132–133° ($CHCl_3$–EtOH), lit [10] mp 133°, $[\alpha]_D$ −17° ($CHCl_3$, 0 84 g/100 ml), IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3520, 1725, 1600, 1560, 1285, 1160, 1115, 1085, 900, 825 ^{1}H NMR ($CDCl_3$, 270 MHz, for comparison with that of **7a**) 6 24 (*d*, *J* = 10 Hz, H-3), 7 63 (*d*, *J* = 10 Hz, H-4), 7 40 (*d*, *J* = 8 Hz, H-5), 6 88 (*d*, *J* = 8 Hz, H-6), 5 32 (*d*, *J* = 9 Hz, H-11), 4 55 (*d*, *J* = 9 Hz, H-12), 4 64 (*quint*, *J* = 1 5 Hz) and 4 60 (*br*, H-14a, b), 1 76 (*br*, H-15) and 3 99 (OMe) Acetylation of 40 mg of murrangatin and prep TLC of the crude product (C_6H_6–EtOAc, 2 1) furnished 30 mg of diacetate **5b**, mp 123–124° (Et_2O), lit [10] mp 124°, IR $\nu_{max}^{CHCl_3}$ cm^{-1} 1735, 1610, 1565, 1285, 1175, 1115, 1025, 900, ^{1}H NMR ($CDCl_3$, 270 MHz, for comparison with that of **7b**) δ 6 25 (*d*, H-3), 7 59 (*d*, H-4), 7 40 (*d*, H-5), 6 84 (*d*, H-6), 6 69 (*d*, H-11), 6 10 (*d*, H-12), 4 91 (*br*) and 4 74 (*quint*, H-14a, b), 1 64 (*br*, H-15), 3 96 (OMe), 2 07, 2 04 (Ac) Benzoylation of 35 mg of murrangatin followed by prep TLC (C_6H_6–EtOAc, 9 1) gave 25 mg of monobenzoate **5c**, mp 198–203° (MeOH–$CHCl_3$), and 15 mg of dibenzoate **5d** mp 100–102° ($CHCl_3$–MeOH), ^{1}H NMR of **5c** ($CDCl_3$, 270 MHz) δ 6 26 (*d*, H-3), 6 86 (*d*, H-6), 6 75 (*d*, H-11), 5 09 (*d*, H-12), 4 82 (*br*) and 4 75 (*quint*, H-14a, b), 1 80 (H-15), 3 98 (OMe), 8 16 (2 arom protons), 7 65–7 36 (H-4, H-5 and three aromatic protons), ^{1}H NMR of **5d** δ 6 26 (*d*, H-3), 6 86 (*d*, H-6), 7 12 (*d*, H-11), 6 35 (*d*, H-12), 5 04 (*br*) and 4 84 (*br*, H-14a, b), 1 81 (H-15), 4 01 (OMe), 8 10 (four aromatic protons), 7 65–7 31 (H-4, H-5 and six aromatic protons), CD curve (MeOH) $[\theta]_{312}$ + 13 900 (max), $[\theta]_{272}$ + 3500 (min), $[\theta]$ + 12 200 (max), $[\theta]_{240}$ 0, $[\theta]_{235}$ −20 800 (min, $\Delta\varepsilon$ − 6 3) $[\theta]_{229}$ 0, $[\theta]_{219}$ + 38 200 (max, $\Delta\varepsilon$ + 11 6)

Hydrolysis of **6c** (a) A soln of 50 mg of **6c** in 5 ml of Me_2CO containing 1 5 ml of 1 N H_2SO_4 was refluxed for 1 hr, diluted with H_2O, extracted with CH_2Cl_2 (5 × 50 ml), concd at red pres and purified by prep TLC (C_6H_6–EtOAc) to give 30 mg of the mixture of **6a** and **6b**, identical with material isolated from the plant (b) A soln of 15 mg of **6c** in 4 ml of MeOH and 1 ml of H_2O containing 50 mg of K_2CO_3 was stirred for 30 min, acidified with HOAc, diluted with H_2O and extracted with $CHCl_3$ The washed and dried extract was concd at red pres , NMR analysis ($CDCl_3$, 270 MHz) of the residue (9 mg) showed that it was a 2 3 mixture of C-14 epimers **6a** and **6b**, signals of **6a** at δ 6 33 (*d*, *J* = 10 Hz, H-3), 7 70 (*d*, *J* = 10 Hz, H-4), 7 46 (H-5), 6 88 (H-8), 5 43 (H-11), 3 79 (H-12), 5 41 (obsc H-14), 1 56 (*br*, H-15) and 3 97 (OMe), signals of **6b** at δ 6 29 (*d*, H-3), 7 70 (*d*, H-4), 7 82 (H-5), 6 87 (H-8), 5 41 (H-11), 3 78 (H-12), 5 52 (*br*, H-14), 1 58 (*br*, H-15) and 3 98 (OMe) [Calc for $C_{15}H_{14}O_6$ MW, 290 0790 Found MW (MS), 290 0774] Other significant ions in the HR-MS were at *m/z* (composition, %) 229 ($C_{13}H_9O_4$, 88) and 213 ($C_{13}H_9O_3$, 100) Reacetylation of this mixture gave material whose NMR spectrum indicated that it was pure **6c**

Oxidation of **6a, b** A soln of 20 mg of **6a, b** in 3 ml of CH_2Cl_2 was stirred with 0 125 g of CrO_3 2 pyridine complex After 5 hr, when reaction was ~ 90% complete (TLC analysis), 2 ml of MeOH was added and then 200 ml of CH_2Cl_2 The washed and

dried soln was evapd, traces of pyridine being removed by co-distillation with toluene, and purified by prep TLC (C_6H_6–EtOAc, 4 1) to yield micromelin (**1**), mp 216–218° (CH_2Cl_2), identical in all respects with material isolated from the plant

Hydrolysis of **8a** A soln of 20 mg of **8a** in 3 ml of EtOH and 5 drops of 10% KOH was stirred at room temp in an N_2 atmosphere for 2 hr at which time starting material had disappeared Dilution with 100 ml of H_2O, acidification with HOAc, extraction with CH_2Cl_2 (5 × 50 ml), evapn of the washed and dried CH_2Cl_2 extracts at red pres, removal of excess HOAc by co-distillation with toluene and purification of the residue by prep TLC (C_6H_6–EtOAc, 2 1) gave 10 mg of a product which NMR analysis (270 MHz, $CDCl_3$) showed to be a 2 1 mixture of 5,7-dihydroxy-6,7,8,4′-tetramethoxyflavone (**9a**) [15] and **5a**, NMR signals of **9a** at δ12 67 (OH), 8 15 (*dm*, J = 9 Hz, H-2′, H-6′), 7 05 (*dm*, J = 9 Hz, H-3′, H-5′), 6 42 (OH) Signals of **5e** at δ6 26 (*d*, J = 10 Hz, H-3), 7 63 (*d*, J = 10 Hz, H-4), 7 40 (*d*, J = 8 Hz, H-5), 6 86 (*d*, J = 8 Hz, H-6), 5 15 (*d*, J = 9 Hz, H-11), 4 90 (*d*, J = 9 Hz, H-12), 4 70 (*br*) and 4 65 (*br*, H-14a, b), 1 71 (*br*, H-15), 3 48 (2H, AB part of ABX_3 system, –O–CH_2Me) and 1 20 (3H, *t*, J = 7 Hz, –OCH_2Me) OMe signals of both compounds were at δ4 06, 4 00, 3 95, 3 93 and 3 88 Acetylation of the mixture (5 mg) followed by usual work-up gave a gummy residue (6 mg) exhibiting two spots which were separated by prep TLC (C_6H_6–EtOAc, 9 1) into **9c** and **8f** which were identified by NMR spectrometry Flavone **9c** had NMR signals ($CDCl_3$) at δ8 12 (*dm*, J = 9 Hz, H-2′, H-6′), 7 06 (*d*, J = 9 Hz, H-3′, H-5′), 4 02, 3 92, 3 88, 3 81 (OMe), 2 52 and 2 44 (Ac), in C_6D_6 at δ8 00 (*dm*, H-2′, H-6′), 6 76 (*dm*, H-3′, H-5′), 3 76, 3 75, 3 62, 3 25 (OMe), 2 34 and 1 89 (Ac) Coumarin **8f** exhibited NMR signals ($CDCl_3$) at δ6 26 (*d*, J = 10 Hz, H-3), 7 65 (*d*, J = 10 Hz, H-4), 7 41 (*d*, J = 8 Hz, H-5), 6 86 (*d*, J = 8 Hz, H-6), 6 09 (*d*, J = 9 Hz, H-11), 5 38 (*d*, J = 9 Hz, H-12), 4 91 (*br*) and 4 68 (*quint*, H-14a, b), 1 58 (*br*, H-15), 3 50 (2H, AB part of ABX_3 system, –OCH_2Me), 1 16 (3H, *t*, J = 7 Hz, –OCH_2Me), 3 98 (OMe) and 2 14 (Ac), MS *m/z* (rel int) 346 (0 6), 233 (75) and 205 (100)

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