*Tetrahedron* Vol. 41, No. 22, pp. 5295 to 5299, 1985
Printed in Great Britain.

0040-4020/85 $3.00 + .00


# CHIRAL SYNTHESIS OF (*R*)-(−)-MELLEIN AND (3*R*,4a*S*)-(+)-RAMULOSIN†

KENJI MORI* and ASHOK K. GUPTA‡
Department of Agricultural Chemistry, The University of Tokyo, Yayoi 1-1-1, Bunkyo-ku, Tokyo 113, Japan

(*Received in Japan* 27 *October* 1984)

**Abstract**—By employing an intramolecular Diels–Alder reaction as the key-step, (*R*)-(−)-mellein **1a** (a metabolite of *Aspergillus melleus*) and (3*R*,4a*S*)-(+)-ramulosin **2** (a metabolite of *Pestalotia ramulosa*) were synthesised from ethyl (*R*)-3-hydroxybutanoate **3a**.

(−)-Mellein **1a** (3-methyl-8-hydroxy-3,4-dihydroisocoumarin) was first isolated by Nishikawa in 1933 as a metabolite of *Aspergillus melleus*.[1] In the same year, Yabuta and Sumiki established the identity of mellein with ochracin, a metabolite of *Aspergillus ochraceus* isolated by them.[2] Since then many workers reported the isolation of mellein from different microbial sources: (+)-mellein from *Fusarium larvarum*,[3] (−)-mellein from *Aspergillus oniki*[4] and also from *Lasiodiplodia theobromae*.[5] Recently (−)-mellein was isolated even as a hairpencil component of the male oriental fruit moth, *Grapholitha molesta*, and found to be involved in the attraction of the female.[6] The proposed gross structure **1a** of mellein was confirmed in 1955 by a synthesis of (±)-mellein methyl ether by Blair and Newbold.[7] We then reported the first synthesis of (±)-mellein itself in 1964.[8] Later, the absolute configuration of (−)-mellein **1a** was proposed to be *R* on the basis of the observation of a negative extremum at 257 nm in its CD spectrum.[9]

(+)-Ramulosin **2** is a metabolite of *Pestalotia ramulosa*§ isolated by Stodola *et al.* in 1964.[10] A recent synthesis of (±)-ramulosin by Cordova and Snider[11] confirmed the proposed structure **2**[10] including the relative stereochemistry as proposed by Tanenbaum *et al.*[12] and by Findlay *et al.*[13] The absolute stereochemistry of (+)-ramulosin as depicted in **2** was proposed by Tanenbaum *et al.*[12] and by Findlay *et al.*[13] considering its apparent biogenetic relationship with (−)-mellein. Actually Tanenbaum *et al.* reported the isolation of (−)-mellein as a metabolite of *Pestalotia ramulosa*.[12]

Almost innumerable different syntheses of (±)-mellein have been reported since our first synthesis was completed. Many of them are based on the so-called directed metallation methodology.[14,15] However, no enantioselective synthesis of (−)-mellein has been reported. Only one existing chiral synthesis yielded (−)-mellein methyl ether of only 75% e.e.[16] As to (+)-ramulosin no attempt has been reported to synthesise it.

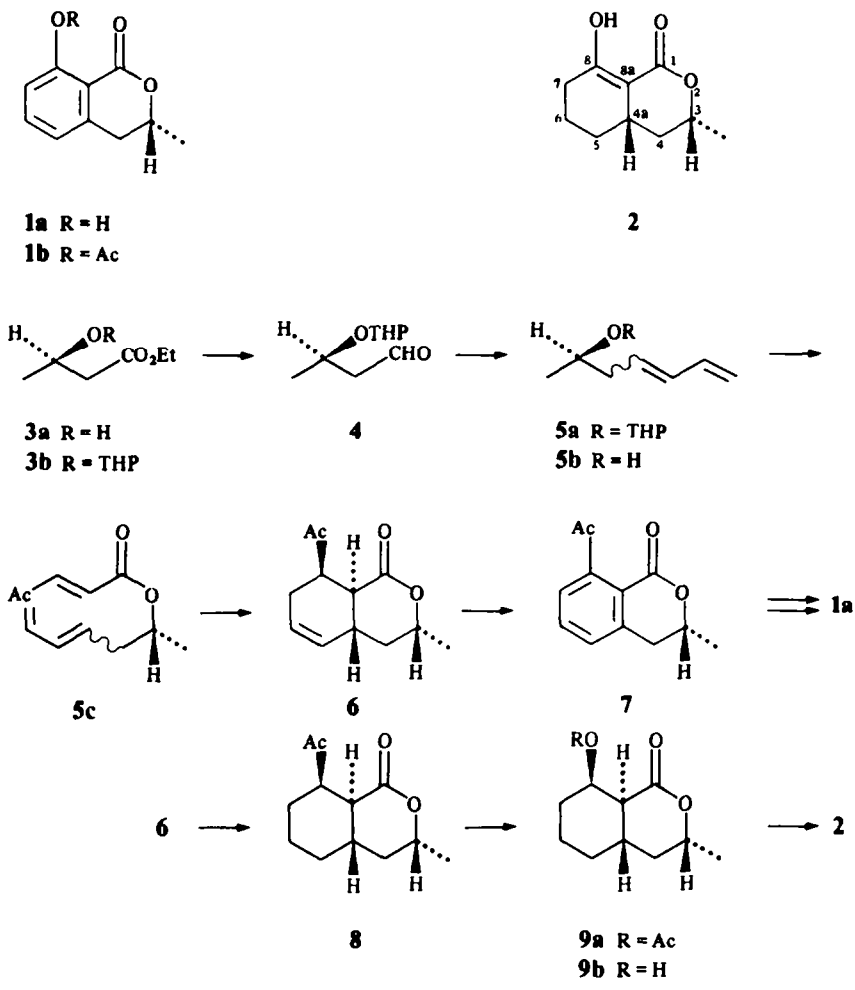
We therefore became interested in synthesising both (−)-mellein **1a** and (+)-ramulosin **2**, so as to conclude our old work.[8] After several unsuccessful attempts to utilise optically active propylene oxide as the chiral source, we developed a route shown in Scheme 1 employing ethyl (*R*)-3-hydroxybutanoate **3a**[17,18] as the starting material. The key-step was the conversion of **5c** to **6** by an intramolecular Diels–Alder reaction.[19,20]

The known THP ether **3b**[18] derived from (*R*)-**3a** was reduced with DIBAL-H to give an aldehyde **4**. (Before carrying out the chiral synthesis described here, a synthesis of (±)-mellein was executed as a model experiment starting from (±)-**3a** (see Experimental).) A Wittig reaction of **4** with $Ph_3P{=}CHCH{=}CH_2$ gave **5a** as a mixture of (*E*)- and (*Z*)-isomers (60:40). After deprotection of the THP group of **5a**, the resulting alcohol **5b** was esterified with dehydrolevulinic acid in the presence of DCC to give an ester **5c**. This was heated in xylene at 140–150° for 6–7 hr to give **6**, m.p. 69–69.5°, $[\alpha]_D^{22}$ +76.5° ($CHCl_3$), in 54% yield. Inspection of its 400 MHz $^1$H-NMR spectrum revealed its stereochemistry as depicted in **6**, since the signal due to C-8a H appeared as dd ($J_{8a,4a}$ = 10 Hz, $J_{8a,8}$ = 3.8 Hz), indicating the presence of an ax Ac group and the more stable *trans* ring-juncture. No other crystalline product could be isolated. The product **6** must have been generated by the less stable initial Diels–Alder adduct. The product **6** with an ax Ac group is more stable than its eq Ac isomer because of the relief of interaction (*peri*-effect) between the eq Ac group and the CO group at C-1. Indeed an attempted isomerisation of **6** by treatment with $K_2CO_3$ in acetone resulted in the recovery of the unchanged **6**. Aromatisation of **6** was effected by successive treatments with NBS and DBU to give **7** in 62% yield. The Baeyer–Villiger oxidation of **7** with $CF_3CO_3H$ gave (*R*)-mellein acetate **1b**. Finally this was hydrolysed with $Et_3N$–MeOH–$H_2O$ to give (*R*)-mellein **1a** as needles, m.p. 55–56°, $[\alpha]_D^{22}$ −100.8° ($CHCl_3$) (lit.[5] m.p. 56°, $[\alpha]_D^{25}$ −102.5° ($CHCl_3$)). Our synthetic **1a** was identical in every respect (IR, NMR, m.p., $[\alpha]_D$) with an authentic sample of (−)-mellein kindly provided by Dr M. Sasaki. No m.p. depression was observed upon admixture of the two samples. Thus (−)-mellein **1a** was synthesised at the Department where it was isolated half a century ago, and its *R*-configuration was established. The overall yield of (−)-**1a** by the present 9-step-synthesis was 7.3% from **3a**.

† In memory of the late Professors T. Yabuta and Y. Sumiki who isolated mellein in 1933 and the gibberellins in 1938. Synthetic Microbial Chemistry—VIII. Part VII, G. Yabuta, Y. Ichikawa, T. Kitahara and K. Mori, *Agric. Biol. Chem.* **49**, 495 (1985).

‡ Postdoctoral fellow (1983–1985) supported by a Monbusho (Japanese Ministry of Education, Science and Culture) scholarship.

§ Cordova and Snider described this as *Pestalotra ramulosa*[11] and Findlay *et al.* as *Pestolatia ramulosa*.[13] These are incorrect and should be read as *Pestalotia ramulosa*.




Scheme 1.

Conversion of the Diels–Alder adduct **6** to (3*R*,4a*S*)-ramulosin **2** was straightforward. Hydrogenation of **6** over Pd–C gave **8**, which was submitted to the Baeyer–Villiger oxidation to furnish **9a**. Treatment of **9a** with *p*-TsOH–MeOH gave a hydroxy lactone **9b**. This was oxidised with the Collins reagent ($CrO_3$–$2C_5H_5N$) in $CH_2Cl_2$ to give (3*R*,4a*S*)-ramulosin **2** as plates, m.p. 118 to 119°, $[\alpha]_D^{22}$ + 18.2° (EtOH) (lit.[10] m.p. 120 to 121°, $[\alpha]_D^{25}$ + 18 ± 2° (EtOH)). The identity of our synthetic **2** with (+)-ramulosin was proved by the mixed m.p. determination by Professor B. Snider. The direct comparison (IR, NMR, m.p., $[\alpha]_D$) of our synthetic sample with an authentic sample of (+)-**2** kindly provided by Dr R. F. Vesonder, U.S.D.A., confirmed the identity of the two samples. This established the absolute configuration of (+)-ramulosin **2** as 3*R*,4a*S*. The overall yield of (+)-**2** by the present 10-step-synthesis was 8.2% from **3a**.

In conclusion both (*R*)-(−)-mellein **1a** and (3*R*,4a*S*)-(+)-ramulosin **2** were synthesised from ethyl (*R*)-3-hydroxybutanoate **3a**.

## EXPERIMENTAL

All b.ps and m.ps were uncorrected. IR spectra were measured as film or as KBr disc on a Jasco A-102 spectrometer. NMR spectra were recorded at 60 MHz with TMS as an internal standard on a Hitachi R-24A spectrometer unless otherwise stated. Optical rotations were measured on a Jasco DIP-140 polarimeter. GLC analyses were performed on a Yanaco G-180 gas chromatograph. Mass spectra were recorded on a Jeolco DX-300 spectrometer at 70 eV.

*Ethyl 3-tetrahydropyranyloxybutanoate* **3b**

Both (*R*)-**3b** and (±)-**3b** were prepared by the known method.[18]

*3-Tetrahydropyranyloxybutanal* **4**

(*a*) (R)-(−)-*Isomer*. A soln of (*R*)-**3b** (16.2 g, 75 mmol) in dry hexane (300 ml) was cooled to −78°. To this was added dropwise DIBAL-H (1.7 M in hexane, 46 ml, 75 mmol) with stirring under Ar at the rate such that the temp did not exceed −70°. After the addition, the mixture was kept at −70° for additional 3 hr. MeOH (1 ml) was then added slowly to the mixture maintaining the temp at −70° for 0.5 hr. A mixture of EtOAc (20 ml) and sat Na · K tartrate (50 ml) was added to the mixture and the temp was allowed to rise to room temp. The mixture was extracted with EtOAc. The EtOAc soln was washed with brine, dried ($Na_2SO_4$) and concentrated *in vacuo*. The residue was distilled to give 12.25 g (95%) of **4**, b.p. 75–76°/10 mm, $n_D^{25}$ 1.4437; $[\alpha]_D^{22}$ − 11.7° (*c* = 1.33, $CHCl_3$); $\nu_{max}$ 2950 (s), 1720 (s) $cm^{-1}$; $\delta$ ($CCl_4$) 1.18 and 1.28 (total 3H, each d, J = 6 Hz), 1.40–1.68 (6H, br), 2.34–2.58 (2H, m), 3.25–4.40 (3H, m), 4.60 (1H, br s), 9.75 (1H, t, J = 2 Hz); MS: *m/z* 172.1057 ($M^+$, calc for $C_9H_{16}O_3$: 172.2236).

(*b*) *Racemate.* In the same manner as described above, (±)-**3b** (16.2 g) yielded 11.7 g (91%) of (±)-**4**, b.p. 74–76°/10 mm, $n_D^{25}$ 1.4575; MS: *m/z* 172 ($M^+$). The IR and NMR spectra were identical with those of (*R*)-**4**.

*4,6-Heptadien-2-ol THP ether* **5a**

(*a*) (R)-(+)-*Isomer.* A soln of n-BuLi in hexane (1.45 M, 51 ml, 74 mmol) was added dropwise to a suspension of $Ph_3PCH_2CH=CH_2 \cdot Br$ (30.6 g, 80 mmol) in dry THF (300 ml) with stirring and ice-cooling at 0–5° under Ar. The stirring was continued for 1 hr under ice-cooling. A soln of (*R*)-**4** (10 g, 60 mmol) in dry THF (20 ml) was added dropwise to the ylid soln over 15 min. The reaction mixture was stirred for 1 hr at 0–5° and for another 1 hr at room temp. It was then filtered and the residue was washed with EtOAc–hexane (1:2). The combined filtrate and washings were concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 5.75 g (50.4%) of **5a**, b.p. 109–110°/10 mm, $n_D^{25}$ 1.4732; $[\alpha]_D^{22}$ + 12.65° (*c* = 0.98, $CHCl_3$); $\nu_{max}$ 1130 (s), 1075 (s), 1020 (s), 1000 (s) $cm^{-1}$; $\delta$ ($CCl_4$), 1.05 and 1.14 (total 3H, each d, J = 6 Hz), 1.30–1.70 (6H, br s), 2.05–2.55 (2H, m), 3.20–4.00 (3H, m), 4.60 (1H, br s), 4.75–7.30 (5H, m). (Calc for $C_{12}H_{20}O_2$: C, 73.43; H, 10.27. Found: C, 73.33; H, 10.21%.)

(*b*) *Racemate.* In the same manner as described above, (±)-**4** (5.16 g) yielded 2.74 g (47%) of (±)-**5a**, $n_D^{25}$ 1.4891; MS: *m/z* 196 ($M^+$). Its IR and NMR spectra were identical with those of (*R*)-**5a**.

*4,6-Heptadien-2-ol* **5b**

(*a*) (R)-(−)-*Isomer.* A soln of (*R*)-**5a** (10 g, 51 mmol) and PPTS (125 mg, 0.5 mmol) in MeOH (40 ml) was stirred for 3 hr at 40–50°. The mixture was diluted with EtOAc (100 ml), washed with 10% $NaHCO_3$ aq, water and brine, dried ($Na_2SO_4$) and concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 4.97 g (87%) of (*R*)-**5b** as a mixture of (*E*)- and (*Z*)-isomers, b.p. 52 to 53°/6 mm, $n_D^{25.5}$ 1.4751; $[\alpha]_D^{22}$ − 6.12° (*c* = 0.98, $CHCl_3$); $\nu_{max}$ 3350 (s), 3100 (w), 1595 (w), 1115 (s), 1000 (s), 900 (s) $cm^{-1}$; $\delta$ ($CCl_4$) 1.10 (3H, d, J = 7 Hz), 2.20 (2H, q, J = 7 Hz), 2.50 (1H, s), 3.70 (1H, q, J = 7 Hz), 4.80–6.85 (5H, m); $^{13}$C-NMR: $\delta$ (25 MHz, $CDCl_3$) 22.83, 37.50, 42.45, 67.35, 67.64, 115.82, 117.89, 127.96, 130.59, 131.88, 132.07, 134.00, 136.87; GLC (column, PEG 20M, 50 m × 0.28 mm at 100°; carrier gas, $N_2$, 1.0 kg/$cm^2$) $R_t$ 6.2 min [61.4%, (*E*)-isomer], 6.5 min (38.6%, (*Z*)-isomer); MS: *m/z* 112.0893 ($M^+$, calc for $C_7H_{12}O$: 112.1712).

(*b*) *Racemate.* In the same manner as described above, (±)-**5a** (2.6 g) yielded 1.45 g (98%) of (±)-**5b**, $n_D^{25}$ 1.4845; GLC (column, PEG 20 M, 50 m × 0.28 mm at 100°; carrier gas, $N_2$, 1 kg/$cm^2$) $R_t$ 6.1 min (58%, (*E*)-isomer), 6.5 min (42%, (*Z*)-isomer); MS: *m/z* 112 ($M^+$), 113 ($M^+$ + 1). Its IR and NMR spectra were identical with those of (*R*)-**5b**.

*Dehydrolevulinic acid*

This was prepared from levulinic acid by the method of Overend *et al.*,[21] m.p. 124–125° (lit.[21] m.p. 125 to 126°), $\nu_{max}$ ~3300 (m), ~2700 (m), 1670 (s), 1620 (m), 1000 (m), 890 (m) $cm^{-1}$; $\delta$ (acetone-$d_6$) 1.85 (3H, s), 6.10 (1H, d, J = 16 Hz), 6.49 (1H, d, J = 16 Hz); $^{13}$C-NMR: $\delta$ (25 MHz, acetone-$d_6$) 28.43, 132.56, 141.54, 167.28, 198.72.

*1-Methyl-3,5-hexadienyl 4-oxo-2-pentenoate* **5c**

(*a*) (R)-(+)-*Isomer.* To a soln of (*R*)-**5b** (3.4 g, 30.3 mmol), DCC (7.5 g, 36.4 mmol) and DMAP (0.3 g) in $CH_2Cl_2$ (60 ml) was added dehydrolevulinic acid (4.8 g, 42.1 mmol). The mixture was stirred for 4 hr at room temp, diluted with hexane (50 ml) and filtered. The solid residue was washed with $CH_2Cl_2$–hexane (1:1, 200 ml). The combined filtrate and washings were concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 5.3 g (84%) of (*R*)-**5c**, b.p. 114–115°/1.5 mm, $n_D^{24.5}$ 1.4896; $[\alpha]_D^{22}$ + 19.2° (*c* = 1.14, $CHCl_3$); $\nu_{max}$ 1720 (s), 1700 (s), 1690 (s), 1640 (w), 1290 (s), 1255 (s) $cm^{-1}$; $\delta$ ($CCl_4$) 1.22 (3H, d, J = 7 Hz), 2.27 (3H, s), 2.20–2.60 (2H, m), 4.70–6.20 (6H, m), 6.42 (1H, d, J = 16 Hz), 6.79 (1H, d, J = 16 Hz). (Calc for $C_{12}H_{16}O_3$: C, 69.21; H, 7.74. Found: C, 68.74; H, 7.72%.)

(*b*) *Racemate.* In the same manner as described above, (±)-**5b** (1.2 g) yielded 1.78 g (80%) of (±)-**5c**, b.p. 110–115°/1.6 mm, $n_D^{25}$ 1.5048; MS: *m/z* 208.1085 ($M^+$, calc for $C_{12}H_{16}O_3$: 208.2446). Its IR and NMR spectra were identical with those of (*R*)-**5c**.

*8-Acetyl-3-methyl-3,4,4a,7,8,8a-hexahydroisocoumarin* **6**

(*a*) (3R,4aR,8R,8aS)-(+)-*Isomer.* A soln of (*R*)-**5c** (3.5 g, 16.8 mmol) in xylene (300 ml) was stirred and heated at 140–150° for 6 to 7 hr. The solvent was removed *in vacuo* and the residue was treated with ether (200 ml). The polymeric by-product was filtered off and washed with ether. The combined ether soln was concentrated *in vacuo*, and the residue was chromatographed over $SiO_2$ to give 1.9 g (54%) of (+)-**6**. This was recrystallised from EtOAc–hexane to give colourless crystals, m.p. 69.0–69.5°, $[\alpha]_D^{22}$ + 76.6° (*c* = 1.08, $CHCl_3$); $\nu_{max}$ 1730 (s), 1720 (s), 1390 (m), 1280 (m), 1210 (m), 1175 (m), 1075 (m), 715 (m) $cm^{-1}$; $\delta$ (400 MHz, $CDCl_3$) 1.24 (1H, ddd, J = 14, 12 and 2.5 Hz), 1.37 (3H, d, J = 6 Hz), 2.22 (3H, s), 2.23 (1H, ddd, J = 14, 9 and 2.5 Hz), 2.39 (1H, dm), 2.62 (1H, dm), 3.02 (1H, m), 3.23 (1H, m), 3.32 (1H, dd, J = 10 and 3.8 Hz), 4.48 (1H, m), 5.50 (1H, dq, J = 10 and 1.5 Hz), 5.69 (1H, m). (Calc for $C_{12}H_{16}O_3$: C, 69.21; H, 7.74. Found: C, 68.99; H, 7.72%.)

(*b*) *Racemate.* In the same manner as described above, (±)-**5c** (1 g) yielded 536 mg (53.6%) of (±)-**6**, m.p. 59.5–60.0°; $\nu_{max}$ 1725 (s), 1710 (s), 1380 (m), 1355 (m), 1260 (m), 1190 (m), 1165 (s), 1120 (m), 1075 (m), 780 (s) $cm^{-1}$. (Calc for $C_{12}H_{16}O_3$: C, 69.21; H, 7.74. Found: C, 68.91; H, 7.50%.) The NMR spectrum of (±)-**6** was identical with that of (*R*)-**6**.

*8-Acetyl-3-methyl-3,4-dihydroisocoumarin* **7**

(*a*) (R)-(−)-*Isomer.* (+)-**6** (1.68 g, 8.0 mmol) was taken in $CCl_4$ (40 ml) and treated with NBS (2.84 g, 16.0 mmol) under reflux for 1 hr. It was then filtered and the residue was washed with EtOAc. The combined filtrate and washings were washed with $NaHCO_3$ aq, water and brine, dried ($Na_2SO_4$) and concentrated *in vacuo* to give 2.5 g of an oil. This was dissolved in dry THF (50 ml) and treated with DBU (2.4 g) under Ar. The soln was stirred for 2 hr at room temp and the excess DBU was neutralised with 10% HCl. The soln was diluted with EtOAc (100 ml), washed with $NaHCO_3$ aq, water and brine, dried ($Na_2SO_4$) and concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 1.02 g (62%) of (−)-**7**. This was recrystallised from EtOAc–hexane to give colourless needles, m.p. 112 to 113°, $[\alpha]_D^{22}$ − 249.3° (*c* = 1.0, $CHCl_3$); $\nu_{max}$ 1710 (s), 1590 (m), 1355 (m), 1270 (s), 1115 (m), 1055 (m), 810 (m), 705 (m) $cm^{-1}$; $\delta$ ($CDCl_3$) 1.50 (3H, d, J = 6 Hz), 2.48 (3H, s), 2.90 (2H, d, J = 7 Hz), 4.65 (1H, q, J = 7 Hz), 7.10 (1H, dd, J = 7 and 2 Hz), 7.20 (1H, dd, J = 6 and 2 Hz), 7.45 (1H, dd, J = 6 and 7 Hz). (Calc for $C_{12}H_{12}O_3$: C, 70.57; H, 5.92. Found: C, 70.21; H, 5.80%.)

(*b*) *Racemate.* In the same manner as described above, (±)-**6** (669 mg) yielded 395 mg (60%) of (±)-**7**, m.p. 115.5–116.0°; $\nu_{max}$ 1700 (s), 1585 (m), 1350 (m), 1275 (s), 1105 (m) $cm^{-1}$. (Calc for $C_{12}H_{12}O_3$: C, 70.57; H, 5.92. Found: C, 70.08; H, 5.93%.) The NMR spectrum of (±)-**7** was identical with that of (*R*)-**7**.

*8-Acetoxy-3-methyl-3,4-dihydroisocoumarin (mellein acetate)* **1b**

(*a*) (R)-(−)-*Isomer.* A soln of $CF_3CO_3H$ in $CH_2Cl_2$ was prepared from $(CF_3CO)_2O$ (2.5 ml, 18 mmol) and 90% $H_2O_2$ (0.4 ml, 15 mmol) in $CH_2Cl_2$ (2 ml). A portion of this soln containing 2.5 mmol of $CF_3CO_3H$ was added slowly over 30 min to a soln of (*R*)-**7** (240 mg, 1.17 mmol) in $CH_2Cl_2$ (5 ml) in the presence of powdered $Na_2HPO_4$ (1.4 g, 10 mmol) at room temp. The mixture was stirred for 10–15 hr at room temp and diluted with EtOAc (50 ml). It was then filtered and the residue was washed with EtOAc. The combined filtrate and washings were washed with $NaHCO_3$ aq, water and brine, dried ($Na_2SO_4$) and concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 120 mg (68%) of (*R*)-**1b**. This was recrystallised from EtOAc–hexane to give colourless needles, m.p. 125–126°, $[\alpha]_D^{22}$ − 170.7° (*c* = 1.01, $CHCl_3$); $\nu_{max}$ 1760 (s), 1715 (s), 1610 (m), 1260 (s), 1200 (s), 1050 (s) $cm^{-1}$; $\delta$

($CDCl_3$) 1.45 (3H, d, J = 6 Hz), 2.32 (3H, s), 2.90 (2H, d, J = 7 Hz), 4.58 (1H, m), 6.95 (1H, d, J = 7.5 Hz), 7.18 (1H, d, J = 6 Hz), 7.42 (1H, dd, J = 6 and 7.5 Hz). (Calc for $C_{12}H_{12}O_4$: C, 65.44; H, 5.49. Found: C, 65.30; H, 5.42%.)

(*b*) *Racemate.* In the same manner as described above, (±)-**7** (50 mg) yielded 25 mg (68%) of (±)-**1b**, m.p. 89–90°, $\nu_{max}$ 1760 (s), 1710 (s), 1610 (m), 1255 (s), 1200 (s), 1050 (s) $cm^{-1}$. (Calc for $C_{12}H_{12}O_4$: C, 65.44; H, 5.49. Found: C, 65.04; H, 5.33%.) The NMR spectrum of (±)-**1b** was identical with that of (*R*)-**1b**.

*Mellein* (8-*hydroxy*-3-*methyl*-3,4-*dihydroisocoumarin*) **1a**

(*a*) (R)-(−)-*Isomer.* A soln of (*R*)-**1b** (85 mg, 0.38 mmol) in $Et_3N$–$H_2O$–MeOH (1 : 1 : 2, 3 ml) was stirred for 2 hr at room temp. It was then diluted with ether (50 ml). The ether soln was washed with 10% HCl, $NaHCO_3$ aq, water and brine, dried ($Na_2SO_4$) and concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 67 mg (97%) of (*R*)-**1a**. Recrystallisation of crude (*R*)-**1a** from hexane gave colourless needles, m.p. 55 to 56°, $[\alpha]_D^{22}$ − 100.8° (*c* = 1.01, $CHCl_3$) (Dr Sasaki's authentic sample showed: m.p. 55 to 56°, $[\alpha]_D^{22}$ − 100.2° (*c* = 1.0, $CHCl_3$); mixed m.p. 55 to 56°); $\nu_{max}$ 3450 (w), 3000 (w), 1665 (s), 1615 (m), 1580 (m), 1460 (s), 1410 (w), 1380 (m), 1365 (m), 1325 (m), 1300 (m), 1235 (s), 1220 (s), 1200 (m), 1165 (m), 1115 (m), 1060 (w), 1050 (m), 955 (m), 900 (w), 810 (s), 785 (w), 740 (m), 700 (m), 680 (m) $cm^{-1}$; δ (100 MHz, $CDCl_3$) 1.54 (3H, d, J = 7 Hz), 2.94 (2H, d, J = 7 Hz), 4.74 (1H, m), 6.68 (1H, dd, J = 6 and 2 Hz), 6.88 (1H, d, J = 7 Hz), 7.38 (1H, dd, J = 7 and 6 Hz); $^{13}$C-NMR (25 MHz, $CDCl_3$) 20.74, 34.60, 76.08, 108.28, 116.18, 117.91, 136.11, 139.38, 162.17, 169.89. (Calc for $C_{10}H_{10}O_3$: C, 67.41; H, 5.66. Found: C, 67.36; H, 5.57%.) The IR, $^1$H-NMR and $^{13}$C-NMR data of our synthetic (*R*)-**1a** were identical with those of the authentic sample.

(*b*) *Racemate.* In the same manner as described above, (±)-**1b** (40 mg) yielded 31 mg (98%) of (±)-**1a**, m.p. 37–38°. Its m.p., IR and NMR spectra were found to be identical with the authentic sample previously synthesised in this laboratory.[8]

(3R,4aS,8R,8aS)-(+)-8-*Acetyl*-3-*methyl*-3,4,4a,5,6,7,8,8a-*octahydroisocoumarin* **8**

A suspension of 10% Pd–C (10 mg) in a soln of (+)-**6** (1.0 g, 4.8 mmol) in EtOAc (50 ml) was stirred under $H_2$ for 3 hr at room temp, when the $H_2$ uptake ceased. It was then filtered and the catalyst was washed with EtOAc. The combined filtrate and washings were concentrated *in vacuo* to give 985 mg (98%) of **8**, b.p. 110–115° (bath temp)/0.3 mm, $n_D^{25}$ 1.4862; $[\alpha]_D^{22}$ + 128° (*c* = 1.2, $CHCl_3$); $\nu_{max}$ 1750 (s), 1710 (s), 1365 (m), 1210 (m), 1120 (m) $cm^{-1}$; δ ($CCl_4$) 1.28 (3H, d, J = 6 Hz), 1.45–2.04 (5H, m), 2.12 (3H, s), 2.14–2.80 (4H, m), 3.07 (2H, m), 4.47 (1H, br); MS: *m/z* 210.1342 ($M^+$, calc for $C_{12}H_{18}O_3$: 210.2604).

(3R,4aS,8R,8aS)-(+)-8-*Acetoxy*-3-*methyl*-3,4,4a,5,6,7,8,8a-*octahydroisocoumarin* **9a**

A soln of $CF_3CO_3H$ (9 mmol) in $CH_2Cl_2$ (3 ml) was prepared in the usual manner as described in the synthesis of **1b**. This was added slowly over a period of 1 hr to a stirred soln of **8** (500 mg, 2.38 mmol) in $CH_2Cl_2$ (10 ml) in the presence of powdered $Na_2HPO_4$ (3.4 g). The mixture was stirred for 15 hr at room temp. It was then filtered and the solid residue was washed with EtOAc. The combined filtrate and washings were washed with $NaHCO_3$ aq, water and brine, dried ($Na_2SO_4$) and concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 376 mg (70%) of **9a**. Recrystallisation of the crude **9a** from EtOAc–hexane gave colourless needles, m.p. 129–130°, $[\alpha]_D^{22}$ + 69.4° (*c* = 1.06, $CHCl_3$); $\nu_{max}$ 1730 (s), 1370 (m), 1235 (s), 1205 (m), 1190 (m), 1110 (m), 1020 (m) $cm^{-1}$; δ ($CDCl_3$) 1.35 (3H, d, J = 6 Hz), 1.40–1.90 (6H, m), 2.06 (3H, s), 2.15–2.90 (4H, m), 4.40 (1H, br m), 5.40 (1H, m). (Calc for $C_{12}H_{18}O_4$: C, 63.69; H, 8.02. Found: C, 63.58; H, 7.85%.)

(3R,4aS,8R,8aS)-(+)-*Hydroxy*-3-*methyl*-3,4,4a,5,6,7,8,8a-*octahydroisocoumarin* **9b**

A soln of **9a** (400 mg, 1.77 mmol) and *p*-TsOH (95 mg, 0.5 mmol) in MeOH (15 ml) was stirred for 18–20 hr at room temp. The solvent was removed *in vacuo* and the residue was taken in EtOAc (100 ml). The EtOAc soln was washed with $NaHCO_3$ aq, water and brine, dried ($Na_2SO_4$) and concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 277 mg (85%) of **9b**. This was recrystallised from EtOAc–hexane to give colourless plates, m.p. 71 to 72°, $[\alpha]_D^{22}$ + 21.3° (*c* = 1.0, $CHCl_3$); $\nu_{max}$ 3450 (s), 1725 (s), 1200 (m), 1100 (m) $cm^{-1}$; δ ($CDCl_3$) 1.35 (3H, d, J = 6 Hz), 1.45–2.65 (10H, m), 3.90–4.60 (2H, m). (Calc for $C_{10}H_{16}O_3$: C, 65.19; H, 8.75. Found: C, 65.38; H, 8.69%.)

(3R,4aS)-(+)-*Ramulosin* (8-*hydroxy*-3-*methyl*-3,4,4a,5,6,7-hexahydroisocoumarin) **2**

$CrO_3$ (1.3 g, 13 mmol) was added to a well stirred and cooled soln of dry pyridine (2.05 g, 26 mmol) in dry $CH_2Cl_2$ (15 ml). The dark soln was stirred for 15 min at room temp. Then a soln of **9b** (240 mg, 1.3 mmol) in dry $CH_2Cl_2$ (2 ml) was added in one portion. A tarry black deposit separated immediately. After stirring for an additional 15 min at room temp, the mixture was diluted with ether (100 ml) and filtered. The residue was washed with ether (50 ml × 3). The combined organic soln was washed with sat $CuSO_4$ aq and brine, dried ($Na_2SO_4$) and concentrated *in vacuo*. The residue was chromatographed over $SiO_2$ to give 185 mg (78%) of **2**. This was recrystallised from EtOAc–hexane to give colourless plates, m.p. 118–119°, $[\alpha]_D^{22}$ + 18.2° (*c* = 1.15, EtOH) (Dr Vesonder's authentic sample: m.p. 118–119°, $[\alpha]_D^{22}$ + 18.3° (*c* = 1.20, EtOH); mixed m.p. 118–119°); $\nu_{max}$ 3430 (m), 2880 (m), 1640 (s), 1615 (sh), 1445 (m), 1405 (m), 1385 (w), 1350 (m), 1300 (m), 1270 (m), 1230 (s), 1195 (m), 1170 (m), 1140 (m), 1105 (m), 1060 (m), 1020 (m), 955 (m), 890 (s), 830 (m), 770 (m) $cm^{-1}$; δ (400 MHz, $CDCl_3$) 1.16 (1H, m), 1.31 (1H, ddd, J = 13, 12.5 and 1.5 Hz), 1.38 (3H, d, J = 6 Hz), 1.60 (1H, s), 1.66 (1H, m), 1.85–1.96 (3H, m), 2.38 (2H, m), 2.51 (1H, br, m); δ (100 MHz, $CDCl_3$) 1.38 (3H, d, J = 7 Hz), 1.45–2.05 (6H, m), 2.30–2.60 (3H, m), 4.46 (1H, ddq, J = 12, 7 and 2.5 Hz); $^{13}$C-NMR: δ (25 MHz, $CDCl_3$) 20.91, 21.73, 29.05, 29.54, 32.97, 37.47, 76.55, 96.82, 171.79, 174.75. (Calc for $C_{10}H_{14}O_3$: C, 65.92; H, 7.74. Found: C, 66.15; H, 7.72%.) The IR, $^1$H-NMR and $^{13}$C-NMR spectra of our synthetic **2** was identical with those of the authentic sample.

*Attempted isomerization of* (+)-**6**

$K_2CO_3$ (10 mg) was added to a soln of (+)-**6** (4 mg) in dry acetone (0.5 ml). The mixture was stirred for 2 hr at room temp and then for 2 hr at 50–55°. The mixture was diluted with ether (10 ml) and filtered. The solid residue was washed with ether. The combined ether soln was concentrated *in vacuo* to give recovered (+)-**6** (4 mg), m.p. 67–68°, whose IR spectrum was identical with that of an authentic (+)-**6**.

*Acknowledgements*—We thank Dr M. Sasaki of Kikkoman Shoyu Co. for the supply of an authentic sample of (−)-mellein. We are grateful to Drs R. F. Vesonder and C. W. Hesseltine, U.S.D.A. Northern Regional Research Center, for the supply of an authentic sample of (+)-ramulosin. We also thank Professor B. Snider, Brandeis University, for carrying out the mixed m.p. determination of ramulosin.

## REFERENCES

[1] H. Nishikawa, *Nippon Nogeikagaku Kaishi* (*J. Agric. Chem. Soc. Jpn*) **9**, 772 (1933).
[2] T. Yabuta and Y. Sumiki, *Ibid.* **9**, 1264 (1933).
[3] J. F. Grove and M. Pople, *J. Chem. Soc. Perkin Trans. I* 2048 (1979).
[4] M. Sasaki, Y. Kaneko, K. Oshita, H. Takamatsu, Y. Asao and T. Yokotsuka, *Agric. Biol. Chem.* **34**, 1296 (1970).
[5] D. C. Aldridge, S. Galt, D. Giles and W. B. Turner, *J. Chem. Soc. (C)* 1623 (1971).
[6] R. Nishida, T. C. Baker, W. L. Roelofs and T. E. Acree, *Abstracts of Papers* PEST 100, 186th A.C.S. National Meeting, Washington, D.C., August 28–September 2 (1983).
[7] J. Blair and G. T. Newbold, *J. Chem. Soc.* 2871 (1955).

[8] M. Matsui, K. Mori and S. Arasaki, *Agric. Biol. Chem.* **28**, 896 (1964).
[9a] H. Arakawa, *Bull. Chem. Soc. Jpn* **41**, 2541 (1968); [b] H. Arakawa, N. Torimoto and Y. Masui, *Liebigs Ann. Chem.* **728**, 152 (1969).
[10] F. H. Stodola, C. Cabot and C. R. Benjamin, *Biochem. J.* **93**, 92 (1964).
[11] R. Cordova and B. B. Snider, *Tetrahedron Letters* **25**, 2945 (1984).
[12] S. W. Tanenbaum, S. C. Agarwal, T. Williams and R. G. Pitcher, *Tetrahedron Letters* 2377 (1970).
[13] J. A. Findlay, J. M. Matsoukas and J. Krepinsky, *Can. J. Chem.* **54**, 3419 (1976).
[14] M. S. Narasimhan and B. H. Bhide, *Tetrahedron* **27**, 6171 (1971).
[15] M. P. Sibi, M. A. Jalil Miah and V. Snieckus, *J. Org. Chem.* **49**, 737 (1984) and refs cited therein.
[16] A. C. Regan and J. Staunton, *J. Chem. Soc. Chem. Commun.* 764 (1983).
[17] T. Sugai, M. Fujita and K. Mori, *Nippon Kagaku Kaishi* (*J. Chem. Soc. Jpn*) 1315 (1983).
[18] K. Mori and H. Watanabe, *Tetrahedron* **40**, 299 (1984).
[19] Review: A. G. Fallis, *Can. J. Chem.* **62**, 183 (1984).
[20] Review: G. Desimoni, G. Tacconi, A. Barco and G. P. Pollini, *Natural Products Synthesis through Pericyclic Reactions*, A.C.S. Monograph 180, pp. 164–191, American Chemical Society, Washington, D.C. (1983).
[21] W. G. Overend, L. M. Turton and L. F. Wiggins, *J. Chem. Soc.* 3500 (1950).