SYNTHESIS OF BRASSINOLIDE, A PLANT GROWTH PROMOTING STEROIDAL LACTONE†

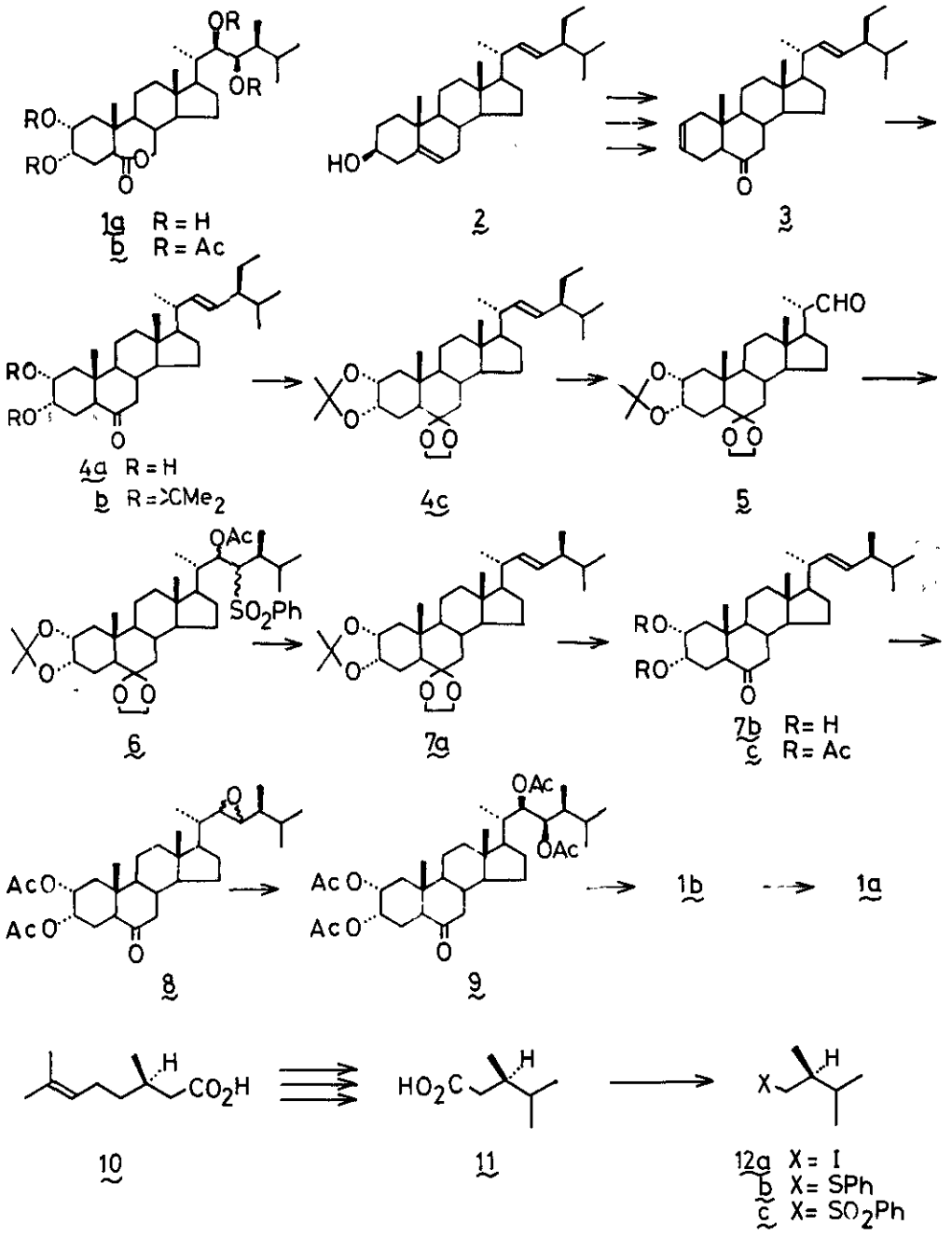
Masayuki Sakakibara, Katsuhide Okada, Yoshitaka Ichikawa and Kenji Mori*
Department of Agricultural Chemistry, The University of Tokyo,
Yayoi 1-1-1, Bunkyo-ku, Tokyo 113, Japan

Abstract — Brassinolide (2α, 3α, 22R, 23R-tetrahydroxy-24S-methyl-B-homo-7-oxa-5α-cholestan-6-one) was synthesized from stigmasterol and (R)-(+)-citronellic acid.

In 1979 a new steroid named brassinolide was isolated (4mg) from 40kg of bee-collected rape pollen (Brassica napus L.) and was assigned the structure 1a by X-ray analysis.[1] The unique structure of brassinolide coupled with its remarkable bioactivity in promoting plant growth aroused interests among synthetic chemists and two syntheses were published.[2,3] We report here another synthesis starting from stigmasterol 2 as an extension of our previous synthesis of (22S, 23S)-homobrassinolide.[4] Similarly to the previous synthesis,[4] the introduction of the two hydroxyl groups on the side chain was executed by the oxidation of the double bond at C-22.

Stigmasterol 2 was converted to a dienone 3 as described previously.[4] This was oxidized with osmium tetroxide and N-methylmorpholine N-oxide in aqueous acetone[5] to give a diol 4a,[6] mp 235-238°; $[\alpha]_D^{21}$ -9.2° ($CHCl_3$), in 97.8% yield. This was converted to the corresponding acetonide 4b, mp 158-159° ; $[\alpha]_D^{24}$ + 21.1° ($CHCl_3$), in quantitative yield by treatment with 2,2-dimethoxypropane and TsOH. After protection (butanone ethylene acetal and TsOH) of the carbonyl group as an ethylene acetal, 4c was treated with ozone. Reductive work-up (dimethyl sulfide in the presence of sodium bicarbonate) of the resulting ozonide yielded an aldehyde 5, mp 118-121° ; $[\alpha]_D^{24.5}$ + 38.2° ($CHCl_3$), in 60% yield from 4b.

† Dedicated to Professor Kyosuke Tsuda on the occasion of his 75th birthday. This work was presented by K.M. as a part of his lecture at the 9th Conference on Isoprenoids in Prague, Czechoslovakia, on September 9, 1981.



Formation of the olefinic side-chain with (24S)-methyl group was accomplished by the Kocienski olefin synthesis[7] employing 5 and a phenyl sulfone 12c. This sulfone 12c was prepared from optically pure (R)-(+)-citronellic acid 10. Conversion of 10 to an acid (R)-11, $[\alpha]_D^{23}$ + 13.7° ($CHCl_3$), was carried out as described

by us in connection with the synthesis of faranal.[8,9] The acid 11 afforded the desired sulfone (R)-(+)-12c, $[\alpha]_D^{23}$ + 19.1° ($CHCl_3$), in 49.2% overall yield from 11 via 12a and 12b by the known method [11 → 12a; I_2/Pb(OAc)$_4$/hν, 12a → 12b: PhSNa, 12b → 12c: MCPBA].[10,11]

Addition of 5 to the carbanion derived from the sulfone 12c was followed by acetylation to give a β-acetoxy sulfone 6. Reduction of 6 with sodium-amalgam in methanol-ethyl acetate (2:1) gave an olefinic product 7a, which upon deprotection furnished a dihydroxy enone 7b, mp 223-227°; $[\alpha]_D^{22}$ + 6.92°($CHCl_3$), in 31% overall yield from 5.[12] The corresponding acetate 7c, mp 195-196°; $[\alpha]_D^{24.5}$ + 3.1°($CHCl_3$), was epoxidized with m-chloroperbenzoic acid to give an epoxide 8, mp 203-204.5°; $[\alpha]_D^{24.5}$ + 16.1° ($CHCl_3$), as a stereoisomeric mixture in 62% yield. The epoxy ring in 8 was cleaved with 30% hydrobromic acid in acetic acid (room temperature, 3 hr) to give a bromo acetate by trans-ring-opening. Another inversion at the carbon bearing the bromine atom was effected by heating with acetic acid-water (4:1) at 100-120° for 19 hr. The product was acetylated with acetic anhydride and 4-(N,N-dimethylamino)pyridine in pyridine to give the desired tetraacetoxy ketone 9, mp 221-224°; $[\alpha]_D^{24.5}$ + 6.81° ($CHCl_3$) [lit.[3] mp 215-217° ; no specific rotation was reported] in 25.3% yield from 8 after chromatographic purification.[13] The Baeyer-Villiger oxidation of 9 with trifluoroperacetic acid in the presence of disodium hydrogen phosphate in methylene chloride yielded brassinolide tetraacetate 1b, mp 218-220°, $[\alpha]_D^{24}$ + 38.96° ($CHCl_3$), in 82.9% yield after chromatographic purification.[14] Hydrolysis of 1b with sodium hydroxide was followed by acidification to give brassinolide 1a, mp 273-275°; $[\alpha]_D^{24}$ + 41.9° ($CHCl_3$-MeOH, 9:1) [lit.[1] mp 274-275°; lit.[2] mp 273-274°; lit.[3] mp 273-278°, $[\alpha]_D^{27}$ + 16° (no specification of the solvent)].[15] The ^{13}C-NMR data of our synthetic brassinolide was in very good accord with those of the natural product.[1]

Full details of this work as well as the synthesis of (22R, 23R)-homobrassinolide and other analogs will be reported in due course.

Acknowledgements — We are grateful to Dr. G. Yabuta, Messrs. H. Ueda, T. Umemura, S. Kuwahara and M. Kondo for their help in the early phase of this work.

REFERENCES AND FOOTNOTES

1. M.D. Grove, G.F. Spencer, W.K. Rohwedder, N. Mandava, J.F. Worley, J.D. Warthen,Jr., G.L. Steffens, J.L. Flippen-Anderson and J.C. Cook,Jr., Nature, 1979, 281, 216.

2. S. Fung and J.B. Siddall, J. Amer. Chem. Soc., 1980, 102, 6580.

3. M. Ishiguro, S. Takatsuto, M. Morisaki and N. Ikekawa, J.C.S. Chem. Comm., 1980, 962.

4. K. Mori, Agric. Biol. Chem., 1980, 44, 1211. The compound described as homobrassinolide in this paper was proved to be the (22S, 23S)-isomer by X-ray analysis.

5. V. VanRheenen, R.C. Kelly and D.Y. Cha, Tetrahedron Letters, 1976, 1973.

6. All new compounds described in this paper gave satisfactory spectral (IR and NMR) and analytical (combustion and/or MS) data.

7. P.J. Kocienski, B. Lythgoe and S. Ruston, J.C.S. Perkin I, 1978, 829.

8. K. Mori and H. Ueda, Tetrahedron Letters, 1981, 22, 1127.

9. The acid (R)-11 was previously obtained either by asymmetric synthesis (82% e.e): T. Mukaiyama, T. Takeda and K. Fujimoto, Bull. Chem. Soc. Japan, 1978, 51, 3368, or by one-carbon elongation of the lower homolog obtained by resolution: P.A. Levene and R.E. Marker, J. Biol. Chem., 1935, 111, 299. Levene and Marker reported the specific rotation of (R)-11 to be $[\alpha]_D^{25}$ +12.8°(neat).

10. P.J. Kocienski, B. Lythgoe and D.A. Roberts, J.C.S. Perkin I, 1978, 834.

11. Kocienski et al.[10] prepared optically impure (S)-(-)-12c, $[\alpha]_D^{40}$-12°($CHCl_3$), starting from (-)-3-methylglutaric half ester obtained by resolution.

12. This olefination reaction is known to give a trans-olefin.[7]

13. Another product was the stereoisomeric (22S, 23S)-tetraacetoxy ketone. This was a non-crystalline gum and easily separated from the desired ketone 9 by chromatography. This stereochemical outcome was the result of double inversion at C-23 or C-24 of the epoxy ring of 8.

14. IR (nujol) 1750 (sh.), 1740 (s), 1722 (s), 1245 (s), 1225 (s), 1050 (m), 1020 (m) cm^{-1}; ^{1}H-NMR (400.5 MHz, $CDCl_3$):δ 0.74 (3H, s), 0.91 (3H, d, J=6.6 Hz), 0.94 (3H,d,J=6.4 Hz), 0.96 (3H, d, J=6.4 Hz), 0.99 (3H, s), 1.01 (3H, d, J=6.8 Hz), 1.19-1.94 (m), 1.996 (3H, s), 2.001 (3H, s), 2.014 (3H, s), 2.110 (3H, s), 2.29 (1H, ddd, J=2.2, 12.4, 15.8Hz), 3.00(1H, dd, J=4.5, 12.1 Hz), 4.05 (1H, dd, J=9.4, 12.5Hz), 4.13 (1H, dd, J=1.2, 12.5 Hz), 4.88 (1H, ddd, J=2.5, 4.4 and 12.5Hz), 5.15 (1H, dd, J=0.4 and 9.3 Hz), 5.33 (1H, dd, J=1.7, 8.8Hz), 5.37 (1H, m).

15. IR (nujol) 3450 (s), 1725 (m), 1693 (s), 1062 (s), 1022 (s), 980 (s), 965 (m) cm^{-1}; ^{1}H-NMR (400.5 MHz, C_5D_5N):δ 0.72 (3H, s), 1.04 (3H, d, J=6.8Hz), 1.05 (3H, s), 1.11 (3H, d, J=6.4Hz), 1.14 (3H, d, J=6.8Hz), 1.21 (3H,d,J=6.3 Hz), 2.31 (1H, dt, J=4.0, 14.5 Hz), 2.52 (1H, ddd, J=2.0, 12.0, 14.0 Hz), 3.60 (1H, dd, J=4.2, 12.0Hz), 3.95 (1H, d, J=8.0 Hz), 3.99-4.11 (3H, m), 4.13 (1H, dd, J=0.5, 8.0Hz), 4.43 (1H, br. s); ^{13}C-NMR (25.0 MHz; CD_2Cl_2-CD_3OD, 9 : 1): δ 10.4, 12.0, 12.2, 15.7, 20.9, 21.1, 68.4, 68.5, 71.3, 73.7, 74.9, 178.1.

Received, 31st August, 1981