SYNTHESIS OF BRASSINOLIDE, A PLANT GROWTH PROMOTING STERoidal LACTONE

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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 1 by X-
ray analysis. The unique structure of brassinolide coupled with its remarkable
bioactivity in promoting plant growth aroused interests among synthetic chemists
and two syntheses were published. We report here another synthesis starting
from stigmasterol 2 as an extension of our previous synthesis of (22S, 23S)-homo-
brassinolide. Similarly to the previous synthesis, 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. This was
oxidized with osmium tetroxide and N-methylmorpholine N-oxide in aqueous acetone
to give a diol 4a, mp 235-238°; [α]D21 -9.2° (CHCl₃), in 97.8% yield. This was
converted to the corresponding acetonide 4b, mp 158-159°; [α]D24 + 21.1° (CHCl₃),
in quantitative yield by treatment with 2,2-dimethoxypropane and TsOH. After pro-
tection (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°; [α]D24.5 + 38.2° (CHCl₃), in 60% yield from 4b.

†Dedicated to Professor Kyosuke Tsuda on the occasion of his 75th birthday.

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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\textsuperscript{7} employing 5 and a phenyl sulfone 12c. This sulfone 12c was prepared from optically pure (R)-(+) -citronelic acid 10. Conversion of 10 to an acid (R)-11, [\alpha]_D\textsuperscript{23} + 13.7° (CHCl\textsubscript{3}), was carried out as described.
by us in connection with the synthesis of faranal. The acid afforded the desired sulfone (R)-(+)\textsubscript{12α}, \([\alpha\textsubscript{D}]+19.1^\circ\text{ (CHCl}_3\text{)}\), in 49.2% overall yield from \textsubscript{11} via \textsubscript{12α} and \textsubscript{12β} by the known method \([\textsubscript{11a} + \textsubscript{11β}; I_2/Pb(OAc)\textsubscript{4}/hv, \textsubscript{11β} + \textsubscript{11α}; PhSNa, \textsubscript{12β} \rightarrow \textsubscript{12α}; MCPBA].\textsuperscript{10,11}

Addition of 5 to the carbanion derived from the sulfone \textsubscript{12α} was followed by acetylation to give a \(\beta\)-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\textdegree; \([\alpha\textsubscript{D}]+6.92^\circ\text{ (CHCl}_3\text{)}\), in 31% overall yield from 5.\textsuperscript{12} The corresponding acetate 7c, mp 195-196\textdegree; \([\alpha\textsubscript{D}]+3.1^\circ\text{ (CHCl}_3\text{)}\), was epoxidized with \(\text{m}\)-chloroperbenzoic acid to give an epoxide 8, mp 203-204.5\textdegree; \([\alpha\textsubscript{D}]+16.1^\circ\text{ (CHCl}_3\text{)}\), 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\textdegree\ 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\textdegree; \([\alpha\textsubscript{D}]+6.81^\circ\text{ (CHCl}_3\text{)}\) [lit.\textsuperscript{3} mp 215-217\textdegree; no specific rotation was reported] in 25.3% yield from 8 after chromatographic purification.\textsuperscript{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\textdegree; \([\alpha\textsubscript{D}]+38.96^\circ\text{ (CHCl}_3\text{)}\), in 82.9% yield after chromatographic purification.\textsuperscript{14} Hydrolysis of 1b with sodium hydroxide was followed by acidification to give brassinolide 1a, mp 273-275\textdegree; \([\alpha\textsubscript{D}]+41.9^\circ\text{ (CHCl}_3\text{-MeOH, 9:1)}\) [lit.\textsuperscript{1} mp 274-275\textdegree; lit.\textsuperscript{2} mp 273-274\textdegree; lit.\textsuperscript{3} mp 273-278\textdegree, \([\alpha\textsubscript{D}]+16^\circ\) (no specification of the solvent)].\textsuperscript{15} The \(^{13}\text{C}-\text{NMR data of our synthetic brassinolide was in very good accord with those of the natural product.}\textsuperscript{1}

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

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REFERENCES AND FOOTNOTES

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.
6. All new compounds described in this paper gave satisfactory spectral (IR and NMR) and analytical (combustion and/or MS) data.
11. Kocienski et al. prepared optically impure (S)-(-)-12G, $[a]_D^{10} -12^\circ$(CHCl$_3$), starting from (-)-3-methylglutaric half ester obtained by resolution.
12. This olefination reaction is known to give a trans-olefin.
13. Another product was the stereoisomeric (22S, 23S)-tetraacetoxy ketone. This was a non-crystalline gum and easily separated from the desired ketone by chromatography. This stereochimical outcome was the result of double inversion at C-23 or C-24 of the epoxy ring of $\Delta^8$.
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$_3$): $\delta$ 0.74 (3H, s), 0.91 (3H, d, J=6.6 Hz), 0.94 (3H, d, J=6.4 Hz), 1.16-1.94 (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, 15.8 Hz), 3.00 (1H, ddd, J=4.5, 12.1 Hz), 4.05 (1H, ddd, J=9.4, 12.5 Hz), 4.13 (1H, ddd, J=1.2, 12.5 Hz), 4.88 (1H, ddd, J=2.5, 4.4 and 12.5 Hz), 5.15 (1H, dd, J=0.4 and 9.3 Hz), 5.33 (1H, dd, J=1.7, 8.8 Hz), 5.37 (1H, m).
15. IR (nujol) 3450 (s), 1725 (m), 1693 (s), 1262 (s), 1022 (s), 980 (s), 965 (m) cm$^{-1}$; $^1$H-NMR (400.5 MHz, CD$_2$D$_2$N): $\delta$ 0.72 (3H, s), 1.04 (3H, d, J=6.8 Hz), 1.05 (3H, s), 1.11 (3H, d, J=6.4 Hz), 1.14 (3H, d, J=6.8 Hz), 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, 12.0 Hz), 3.95 (1H, d, J=8.0 Hz), 3.99-4.11 (3H, m), 4.13 (1H, dd, J=0.5, 8.0 Hz), 4.43 (1H, br. s) $^{13}$C-NMR (25.0 MHz; CD$_2$Cl$_2$-CD$_3$OD, $\delta$): 10.4, 12.0, 12.2, 15.7, 20.9, 21.1, 68.4, 68.5, 71.3, 73.7, 74.9, 178.1.

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