Chemical Conversion of Gibberellin A₁₃ to Gibberellin A₄

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Gibberellin A₁₃ was converted into gibberellin A₄ through oxidative decarboxylation of C-10 carboxyl with lead tetraacetate followed by C-4→C-10 lactone formation.

In the biosynthesis of gibberellins one of the most interesting parts lies in the conversion of C₂₀ gibberellins to C₁₉ gibberellins by the loss of one carbon substituent at C-10 on the *ent*-gibberellane skeleton. Although several hypotheses were proposed concerning the mechanism of this part of biosynthesis, any of them have not been definitely confirmed so far. On the other hand, chemical conversion of C₂₀ gibberellins to C₁₉ gibberellins is not only interesting from the synthetical point of view but also very significant in structural correlation of gibberellins; that is, structures of C₁₉ gibberellins have been definitely established by the correlation to a gibberellin A₃ (GA₃) derivative whose structure was decided by X ray analysis, while structures of C₂₀ gibberellins have been determined mainly by spectral analyses. We succeeded in the chemical conversion of GA₁₃ to GA₄.^{*} Recently Bearder *et al.* reported the chemical conversion of GA₁₃ to GA₄ and its isomeric lactone through oxidative decarboxylation with lead tetraacetate.¹⁾ This prompted us to report our conversion in details.

GA₁₃ (I) was converted to an anhydride (III) according to the method reported by Glat.²⁾ The methyl ester (IV) of III, which was obtained by diazomethane treatment, was treated with sodium methoxide in methanol to afford a diester monoacid (II). The presence of a carboxyl group at C-10 in II was deduced from

the result in methanolysis of a GA₁₃ norketone.³⁾ Since oxidative decarboxylation of the compound XV with lead tetraacetate has been known to give a trisubstituted olefin as a main product,⁴⁾ II was treated with lead tetraacetate in boiling benzene to remove the C-10 substituent by oxidative decarboxylation. The product was found to be decarboxylated ester by mass spectral analysis but NMR spectrum and gas chromatography indicated that the product was a mixture of three isomeric compounds. The main component of the mixture was confirmed to be identical with an ester (V), which can be prepared from a hydrogenolysis product (VII) of GA₇ methyl ester (XIII), by comparison of gas chromatograms, NMR and mass spectra. The structures of other two components were not determined definitely but they are probably a C-5~C-10 and a C-9~C-10 olefinic isomer. As the model experiment to form a C-4→C-10 lactone, V prepared from GA₇ methyl ester (XIII) was hydrolysed and the resultant acid (VI) was treated with iodine in methylene chloride-tetrahydrofuran-aqueous sodium bicarbonate mixture to afford an iodolactone (IX), according to the method used in elaboration of the ring A system of GA₃ by Corey *et al.*⁵⁾ Since separation of components in decarboxylated mixture obtained from II was difficult, the mixture was hydrolysed without separation, and the hydrolysis product containing VI was subjected to the same procedure as in the model experiment. In this case only VI was lactonized and the iodolactone (IX) was isolated, as expected, by preparative

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I $R^1=R^2=R^3=R^4=H$
 II $R^1=Ac, R^2=R^3=CH_3, R^4=H$

III $R=H$
 IV $R=CH_3$

V $R^1=Ac, R^2=R^3=CH_3, R^4=H$
 VI $R^1=R^2=R^3=R^4=H$
 VII $R^1=R^2=R^4=H, R^3=CH_3$
 VIII $R^1=R^2=H, R^3=CH_3, R^4=OH$

IX $R^1=I, R^2=R^3=H$
 X $R^1=R^2=R^3=H$
 XI $R^1=R^2=H, R^3=OH$
 XII $R^1=I, R^2=CH_3, R^3=OH$

XIII $R=H$
 XIV $R=OH$

XV

thin-layer chromatography (TLC). IX was reduced with sodium borohydride in dimethyl sulfoxide,⁶⁾ an iodine at C-1 being replaced by a hydrogen. $GA_4(X)$ thus obtained contained about 13% of an isomer containing an endocyclic olefin in ring D, which arises easily by allylic rearrangement from C-13 deoxygibberellins during reaction and separation process. The endocyclic olefinic isomer was removed by TLC using silica gel impregnated with silver nitrate⁷⁾ and the purified GA_4 was found to be identical with authentic specimen by melting point, infrared and mass spectrum.

Lactone formation between C-4 carboxyl and C-10 in C-1~C-10 double bond with iodine followed by sodium borohydride reduction constitutes the useful method for the preparation of GA_1 . Thus, hydrogenolysis product (VIII) of GA_3 methyl ester (XIV), which is obtained as main product by partial hydrogenation, was converted into GA_1 (XI) by the method in appreciable yield.

EXPERIMENTAL

IR, NMR and mass spectrum were determined with a JASCO IR-KCl spectrometer, a JEOL MH-100 spectrometer and a Hitachi RMU-6L spectrometer, respectively. Melting points (uncorrected) were determined by a Yanaco micro melting point apparatus. Gas chromatography was carried out by a Hitachi K53 gas chromatograph equipped with a flame ionization detector. Carrier gas was N_2 . Columns, 1 m length and 3 mm diameter, packed with 3% SE-30 or 3% XE-60. Temperature, 197°C. TLC was carried out by use of silica gel GF₂₅₄ (Merck) and developing solvent; (a) ethyl ether-*n*-hexane (1:1, v/v), (b) benzene-acetone-acetic acid (14:5:1, v/v), (c) *n*-hexane-ethyl acetate-chloroform-acetic acid (30:20:8:1, v/v).

Preparation of anhydride (III)

GA_{13} (I, 958 mg) was converted to the anhydride (III) by a method in the literature.²⁾ Yield, 750 mg. Mp, 264~268°C. IR ν_{max}^{NaJol} cm^{-1} : 3180, 1797, 1760, 1748, 1721, 1670, 1650, 897. MS m/e : 402 (M^+), 356, 314, 296, 232.

The methyl ester (IV) of III

III was dissolved in methanol and methylated with

diazomethane to afford a solid ester (IV). IR $\nu_{\text{max}}^{\text{Nujol}}$ cm⁻¹: 1804, 1770, 1742, 1659. NMR (in CDCl₃) σ : 1.16 (3H, s), 2.13 (3H, s), 2.45 (1H, d, $J=11$ Hz), 2.76 (1H, d, $J=11$ Hz), 3.72 (3H, s), 4.93 (2H, broad s), 5.08 (1H, broad s). MS m/e : 416 (M⁺), 385, 370, 356, 284.

Methanolysis of anhydride (IV)

IV (740 mg) was dissolved in chloroform (10 ml) and 2 molar equivalents of sodium methoxide (0.1 N methanol solution) was added to it and the solution was kept at 0°C. After 5 min 100 ml of water was added to the solution, which was acidified with 1 N H₂SO₄ (20 ml) and extracted with ethyl acetate. After removal of the solvent, the extract was roughly separated by use of a silicic acid column and eluting solvent of ethyl acetate-benzene, and then purified by TLC using solvent system *c* to afford diester monoacid (II, 430 mg). Mp, 218~220°C. IR $\nu_{\text{max}}^{\text{Nujol}}$ cm⁻¹: 3200, 1731, 1722, 1690, 1651. MS m/e : 448 (M⁺), 430, 416, 388, 370, 328.

Oxidative decarboxylation of II

II (229 mg) was dissolved in dry benzene (25 ml) and to the solution were added cupric acetate (50 mg) and lead tetraacetate (400 mg). The solution was refluxed under nitrogen stream for 15 hr. The benzene solution was poured onto aqueous FeSO₄ solution and the mixture was extracted with diethyl ether. After evaporation of solvent the extract was purified by preparative TLC using solvent system *a* to afford a solid mixture (124 mg). The mixture consisted of three components and the main one was 58% of the whole mixture based on gas chromatography.

Preparation of V from GA₇ methyl ester (XIII)

GA₇ methyl ester was hydrogenated according to the method reported by Cross *et al.*⁸⁾ The resultant hydrogenolysis product (VII) was methylated with diazomethane and acetylated with pyridine and acetic anhydride to afford V as a non-crystalline solid. IR $\nu_{\text{max}}^{\text{Nujol}}$ cm⁻¹: 1729, 1668, 890. NMR (in CDCl₃) σ : 1.20 (3H, s), 2.05 (3H, s), 3.06 (1H, s), 3.62 (3H, s), 3.67 (3H, s), 4.87 (2H, broad s), 5.22 (1H, broad s), 5.27 (1H, broad s). MS m/e : 402 (M⁺), 370, 342, 327, 310, 282.

Hydrolysis of V

V (172 mg) was dissolved in ethanol (0.5 ml) and 1.5 N NaOH aq. solution was added. The solution was kept at 100°C for 6 hr and then extracted with ethyl acetate at pH 3. After drying over Na₂SO₄, evaporation of solvent gave diacid (VI, 67 mg) as solid. IR $\nu_{\text{max}}^{\text{Nujol}}$ cm⁻¹: 3200, 2480, 1700, 886. MS m/e : 332 (M⁺), 314, 296, 286, 270, 268.

The decarboxylated mixture from III was hydrolysed

in the same manner as in the case of V to afford 51 mg of an acidic mixture.

Iodolactonization of decarboxylated product

The hydrolysate of the decarboxylated mixture (51 mg) containing VI was dissolved in tetrahydrofuran (1.5 ml) and methylene chloride (0.5 ml). To the solution were added aqueous solution of NaHCO₃ (5%, 0.2 ml) and iodine (16 mg), and the mixture was kept at room temperature with stirring. After 16 hr ethyl acetate was added to the solution and the organic layer was washed four times with water of pH 2. After drying over Na₂SO₄ evaporation of the solvent gave solid (68 mg), which was purified by TLC (solvent system *b*) to afford iodolactone (IX, 31 mg). Mp, 98~100°C. IR $\nu_{\text{max}}^{\text{Nujol}}$ cm⁻¹: 3500, 1760, 1700, 1645. NMR (in CDCl₃) σ : 1.25 (3H, s), 3.76 (1H, d, $J=11$ Hz), 3.98 (1H, broad s), 4.39 (1H, d, $J=5$ Hz), 4.89 (1H, broad s), 4.97 (1H, broad s). MS (the methyl ester) m/e : 472 (M⁺), 440, 412, 352.

Sodium borohydride reduction of iodolactone (IX)

IX (27 mg) was dissolved in dimethyl sulfoxide (4 ml) and NaBH₄ (60 mg) was added to the solution. The solution was kept at 80°C for 1 hr with stirring. After worked up as usual, the reduction product was obtained as solid, which was purified by TLC (solvent system *b*) to afford crystal of GA₄ (X, 9.3 mg). The endocyclic olefinic isomer contained in the product was removed by use of thin layer of silica gel impregnated with AgNO₃. GA₄ thus obtained was identical with an authentic specimen.

Preparation of GA₁ (XI) from hydrogenolysis product (VIII) of GA₃ methyl ester (XIV)

VIII (200 mg), which was obtained from GA₃ methyl ester as a main product (over 70% yield) by partial hydrogenation as in the case of GA₇ methyl ester, was dissolved in tetrahydrofuran (6 ml) and methylene chloride (2 ml). To the solution were added aq. 5% NaHCO₃ solution (1 ml) and iodine (77 mg), the solution being kept at room temperature with stirring. After 4 hr the product was extracted and purified as in the case of IX. On crystallization from acetone-*n*-hexane an iodolactone (XII) was obtained as crystalline rods. Yield, 237 mg. Mp. 209~210°C. IR $\nu_{\text{max}}^{\text{Nujol}}$ cm⁻¹: 3460, 3320, 1754, 1738, 1657, 900. NMR (in CDCl₃) σ : 1.21 (3H, s), 2.79 (1H, d, $J=10$ Hz), 3.77 (3H, s), 3.85 (1H, d, $J=10$ Hz), 3.99 (1H, m), 4.42 (1H, d, $J=6$ Hz), 5.01 (1H, broad s), 5.29 (1H, broad s). MS m/e : 488 (M⁺), 456, 429, 361, 343, 329, 302.

Sodium borohydride reduction of XII

XII (200 mg) was dissolved in dimethyl sulfoxide (10 ml) and NaBH₄ (200 mg) was added to the solution. The solution was kept at room temperature with

stirring. After 16 hr reduction product was extracted and purified as in the case of IX. The crystal thus obtained was confirmed to be GA₁ methyl ester by IR and mass spectrum. Yield, 81 mg. GA₁ methyl ester thus obtained was converted to GA₁ (XI) by the method of Bartlet *et al.*⁹⁾

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