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Structures of Gibberellins A₃₃ and A₃₄ from Immature Seeds of *Calonyction aculeatum*

Sir:

The isolation of gibberellins from immature seeds of evening-glory (*Calonyction aculeatum*) and the structural determination of two new gibberellins, GA₃₀ and GA₃₁, have been previously reported.¹⁾ Here we report the structural determination of other two new gibberellins, GA₃₃ and GA₃₄.

From 23 kg of immature seeds of evening-glory GA₃₃ (mp 219~221°C) and GA₃₄ (mp 218~219°C) were obtained in yields of 17 mg and 12 mg, respectively, through the procedures already described.¹⁾ On treatment with diazomethane GA₃₃ affords a methyl ester, the high resolution mass spectrum of which shows its molecular formula to be C₂₀H₂₄O₇. The ester also reveals M-18, M-32, M-44, M-46, M-50, M-60, M-78 and M-122 peaks, which constitute a pattern characteristic of C₁₉ gibberellin methyl esters.²⁾ Since the presence of a 3H singlet due to a methoxycarbonyl in the NMR spectrum of the methyl ester shows GA₃₃ to be a monocarboxylic acid, the molecular formula C₁₉H₂₂O₇ is assigned to GA₃₃. The functional groups in GA₃₃ are summarized in Table I. The presence of an α -oriented hydroxyl group at C-12 in *ent*-gibberellane skeleton (I) is shown by a 1H triplet at τ 6.26 in NMR spectrum measured in deuteroacetone as in the cases of GA₃₀ and GA₃₁. An oxidation product of GA₃₃ obtained by the Jones reagent shows a UV absorption maximum at 283 m μ

TABLE I. FUNCTIONAL GROUPS OF GA₃₃

IR (cm ⁻¹ , nujol)	NMR (τ , in deuteroacetone)
2 H-C-CH	3470 5.58 (1H, d), 6.26 (1H, t)
1 COOH	1696
1 >C=O	1733
1 γ -lactone	1794
1 >C=CH ₂	1658 5.02 (1H, s), 4.92 (1H, s)
1 -C-CH ₃	8.89 (3H, s)
1 -CO-CH ₂ - CH(OH)-	7.46 (1H, d), 7.06 (1H, q)

(ϵ 360), which is characteristic of β , γ -unsaturated ketones as in GA₂₆, the presence of the C-12 hydroxyl group being confirmed. In the NMR spectrum of GA₃₃ there exist a 1H doublet at τ 7.46 and a 1H quartet at τ 7.06, which are geminally coupled with one another (J =17 cps), indicating the presence of a methylene group adjacent to a carbonyl. The proton, to which the latter signal is assigned, is further coupled with a carbinyl proton (τ 5.58, J =5 cps). These evidences indicate the presence of the partial structure -CO-CH₂-CH(OH)-, which must be contained in the ring A. Further, the low τ value of the C-5 proton (τ 6.52), which is coupled with the C-6 proton (J =11 cps) to constitute a pattern characteristic of gibberellins, indicates that a hydroxyl exists at C-1 or C-3 in β configuration, suggesting possible two structures II and III for GA₃₃. Structure II was chosen of the two alternatives on the following basis. A hydrogenolysis product (IV) of GA₃ methyl ester was methylated with diazomethane and then oxidized with the Jones

1) N. Murofushi, T. Yokota and N. Takahashi, *Agr. Biol. Chem.*, **34**, 1436 (1970).

2) N. Takahashi, N. Murofushi, S. Tamura, N. Wasada, H. Hoshino, T. Tsuchiya, S. Sasaki, T. Aoyama and E. Watanabe, *Org. Mass Spectrometry*, **2**, 711 (1969).

reagent to afford a keto-ester (V). The ester was oxidized with *m*-chloroperbenzoic acid to afford an epoxide (VI). Treatment of VI with dilute sulfuric acid resulted in cleavage of the epoxide and simultaneous lactonization to give VII. The signals of C-1, C-2, C-5, C-6 and C-18 protons in the NMR spectrum of VII is quite similar to those of GA₃₃ methyl ester; VII and GA₃₃ show an identical CD curve with positive Cotton effect at 296 $m\mu$ ($\theta=10,000$). The other spectral data are in good agreement with the stereochemical expression of II for GA₃₃.

In the same way as in GA₃₃, the molecular formulas, C₁₉H₂₄O₆ and C₂₀H₂₆O₆, are assigned to GA₃₄ and its methyl ester, respectively; their spectral properties show that GA₃₄ is a C₁₉-gibberellin. The functional groups of GA₃₄

are summarized in Table II. The presence of two hydroxyl groups in GA₃₄ is confirmed

TABLE II. FUNCTIONAL GROUPS OF GA₃₄

IR (cm ⁻¹ , nujol)		NMR (τ , in deuteroacetone)	
2	H-C-OH	3340	6.25 (1H, m), 6.38 (1H, d)
1	COOH	1728 ^{a)}	
1	γ -lactone	1776 ^{a)}	
1	$>C=CH_2$	1655	5.16 (1H, s), 5.04 (1H, s)
1	-C-CH ₃		8.85 (3H, s)

^{a)} Determined in dioxane.

by the mass spectrum of the trimethylsilyl ether of its methyl ester (parent peak, *m/e* 506). In the NMR spectrum (in deuteroace-

tone) a 3H singlet due to the C-18 methyl protons and a 1H doublet due to the C-5 proton are observed at τ 8.85 and τ 6.75, respectively. These signals shift to τ 8.29 and τ 6.02, respectively when measured in deuteropyridine, indicating that one of the hydroxyl groups is located at C-3 in β axial. The presence of 1,2-diol is shown by rapid consumption of periodate and by easy formation of an acetonide derivative of GA₃₄ methyl ester. The NMR pattern in the carbonyl region is quite similar to those of GA₈, GA₂₆ and GA₂₇. Since the C-3 hydroxyl group is in β axial, the C-2 hydroxyl group must be in β equatorial. Thus, structure VIII was proposed to GA₃₄.

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