

STRUCTURES OF GIBBERELLINS A₂₆ AND A₂₇ IN IMMATURE SEEDS OF PHARBITIS NIL

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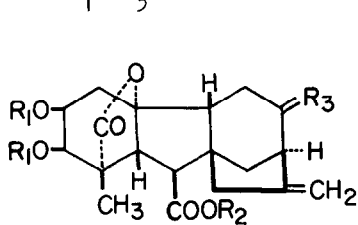
Isolation and biological activities of new gibberellins A₂₆ and A₂₇ in immature seeds of the Japanese morning-glory (Pharbitis nil) have been reported (1). We wish to report here the elucidation of their structures.

Gibberellin A₂₆ (GA₂₆), m.p. 254-257°, formed a monomethyl ester (II), m.p. 207-211°. The high resolution mass spectrum of II reveals a parent ion peak at m/e 376.159 with the composition C₂₀H₂₄O₇ (calcd. 376.152). Therefore GA₂₆ has the molecular formula C₁₉H₂₂O₇. The significant peaks at M-32 (CH₄O) and M-60 (C₂H₄O₂) in the mass spectrum of II, together with an AB quartet due to the C-10, C-10a protons centered at τ 7.27 and τ 6.63 (J=10cps) in its NMR spectrum (CDCl₃), suggest that GA₂₆ retains the structural features common to the known gibberellins.

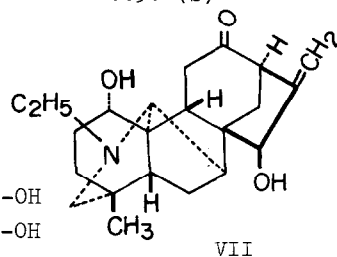
As summarized in Table I, the functional groups of II are deduced on the basis of the IR, UV and NMR spectra. GA₂₆ rapidly consumed periodate and easily formed a diacetate (III), m.p. 209-210°, having no free hydroxyl group. These suggest the presence of a cis-1,2-glycol system in GA₂₆. An axial hydroxyl at C-2 is indicated by the down-field shifts of the C-1 methyl singlet (τ 8.79 in CDCl₃; τ 8.50 in C₅D₅N) and the C-10a proton doublet (τ 6.63 in CDCl₃; 6.09 in C₅D₅N) in the change of the solvent from CDCl₃ to C₅D₅N (2). Therefore the other hydroxyl must be attached to C-3 in the equatorial conformation. The C-2 proton doublet at τ 6.25 (J=4.0 cps) rationalizes the existence of a hydroxyl substituent at C-3 and the equatorial conformation of the C-3 hydroxyl is supported by the C-3 proton multiplet with a large width (20 cps) characteristic of an axial proton. The pattern of $\begin{array}{c} | \\ \text{H}-\text{C}-\text{OH} \\ | \end{array}$ region is very similar to that of GA₈. Therefore GA₂₆ contains a C-2_a, C-3_e-glycol.

Table I. Functional Groups in GA₂₆ Methyl Ester

	IR, cm ⁻¹	NMR, τ(ppm)	
	(CHCl ₃)	(CDCl ₃)	(C ₅ D ₅ N)
2 OH	3500	6.13(m), 6.25(d)	5.70(m), 5.85(d)
		($\underset{ }{\text{H}}\text{C}-\text{OH}$)	
1 γ-lactone	1780		
1-COOCH ₃	1733	6.29 (s)	6.34 (s)
1 >C=O	1716		
1 >C=CH ₂	1655	4.80, 4.95 (broad s)	4.75, 5.00 (broad s)
1 $\text{-}\underset{ }{\text{C}}\text{-CH}_3$		8.79 (s)	8.50 (s)



- I R₁=R₂=H, R₃=0
- II R₁=H, R₂=CH₃, R₃=0
- III R₁=Ac, R₂=H, R₃=0
- IV R₁=Ac, R₂=CH₃, R₃=0
- V R₁=Ac, R₂=CH₃, R₃=H, β-OH
- VI R₁=Ac, R₂=CH₃, R₃=H, α-OH



II shows a UV absorption at λ_{max} 288 mμ (ϵ 280) and a CD curve having a negative maximum at λ 294 mμ (Θ -8.97×10^3), which are characteristic of β,γ-unsaturated ketones in the appropriate molecular geometry (3). Since GA₂₆ is considered to retain the structural features common to gibberellins, the position of the ketone should be limited either at C-6 or C-11. The IR band at 1716 cm⁻¹ (CHCl₃) is indicative of a six-membered ring ketone. NaBH₄ reduction of a diacetate of II (IV) afforded an amorphous isomeric mixture of V and VI (4:1), which could be separated on TLC. The NMR spectra of V and VI show new multiplets* of the type $\underset{|}{\text{H}}\text{-}\underset{|}{\text{C}}\text{-OH}$ at τ 5.88 (W=23 cps) and τ 6.25, respectively. This establishes the position of the ketone at C-6, since an alcohol from a C-11 ketone is expected to give a C-11 proton doublet.

An unusual broad doublet at τ 6.77 (W=10 cps) in the NMR spectrum of IV can be assigned to the C-7 proton, which shifts to the up-field (τ 7.16, W=18 cps) in that of V.

All evidences above cited as well as the consideration of the structural

* These τ values showed that V has an equatorial C-11 proton and VI has an axial C-11 proton because an equatorial proton resonates at lower field than an axial one.

relationship to other members of the gibberellin group allow to assign the structure I for GA₂₆.

It is further supported by the following observation. An aconite alkaloid, songorine (VII) (4), whose structure has been decided as VII by X-ray analysis, possesses the same structure and stereochemistry of the C,D rings as GA₂₆. The CD curve of VII (a negative maximum at 298 mμ, $\ominus -1.05 \times 10^4$) is quite similar to that of GA₂₆. This furnishes the strong support not only for the position of the ketone but also the stereochemistry of the C,D rings.

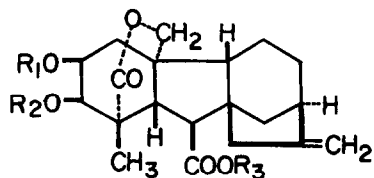
GA₂₇, m.p. 163-165° (hydrate), was converted to a monomethyl ester (IX), m.p. 181-183°. The molecular formula C₂₀H₂₆O₆ for GA₂₇ was established by the high resolution mass spectrum of IX (a parent ion peak at m/e 376.186; calcd. for C₂₁H₂₈O₆, 376.189). Assignments of the NMR and IR spectra of IX for functional groups are illustrated in Table II. The presence of these functional groups in IX as well as the mass spectral features [M-32 (CH₄O) and M-60 (C₂H₄O₂)] shows GA₂₇ to be a new C₂₀ gibberellin.

GA₂₇ rapidly consumed periodate. It readily formed a diacetate (X), m.p. 226-228°, and an isopropylidene derivative (XI), m.p. 216-217°, both of which lack free hydroxyls. These suggest the presence of a cis-1,2-glycol system in GA₂₇. In the similar manner to that for GA₂₆, GA₂₇ is shown to have a C-2_a,C-3_e-glycol.

The presence of a δ -lactone is indicated by the IR band at 1740 or 1710 cm⁻¹ (nujol) and an AB quartet centered at τ 5.54 and 5.88 (J=13 cps) in the NMR spectrum (CDCl₃). The similar NMR patterns have been reported for GA₁₅ (5) and 2,7-dihydroxy-GA₁₅ methyl esters (6) containing δ -lactones between C-1 and C-4a.

Table II. Functional Groups in GA₂₇ Methyl Ester

	IR, cm ⁻¹ (nujol)	NMR, τ (ppm)	
		CDCl ₃	C ₅ D ₅ N
2 OH	3300	6.14 (m), 6.28 (d) ($\begin{array}{c} \\ \text{H}-\text{C}-\text{OH} \\ \end{array}$)	5.65 (m), 5.82 (d)
1 -COOCH ₂ - (δ -lactone)	1740, 1710	5.54, 5.88 (AB q) (-COO- $\begin{array}{c} \\ \text{CH}_2 \\ \end{array}$ -)	5.42, 5.81 (AB q)
1 -COOCH ₃		6.31 (s)	6.37 (s)
1 >C=CH ₂	1650	5.05, 5.17 (broad s)	5.07, 5.21 (broad s)
1 $\begin{array}{c} \\ -\text{C}-\text{CH}_3 \\ \end{array}$		8.76 (s)	8.31



- VIII $R_1=R_2=R_3=H$
 IX $R_1=R_2=H, R_3=CH_3$
 X $R_1=R_2=Ac, R_3=CH_3$
 XI $R_1, R_2=(CH_3)_2C<, R_3=CH_3$

A signal due to the C-10, C-10a protons, characteristic of the gibbane skeleton, is found as a 2H singlet at τ 7.24 in the NMR spectrum ($CDCl_3$) of IX, while in C_5D_5N it is splitted into an AB quartet centered at τ 6.73 and 6.96 ($J=13$ cps). The similar NMR patterns have been observed in the spectra of GA_{22} (7) and 2,7-dihydroxy- GA_{15} methyl esters.

The structure VIII for GA_{27} accomodates all evidences above cited.

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