21. *Gibberellic Acid. Part XX.* Stereochemistry of Ring* A.

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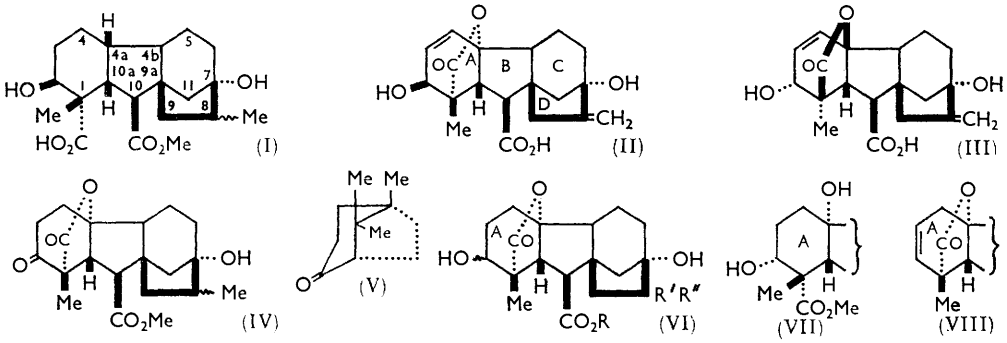
Gibberellic acid is shown to have the absolute configuration (II) with the ring A lactone bridge in the α-configuration.

Ring D in gibberellic acid (II) has been shown to have the β(absolute)-configuration.[1] The stability to acid at position 10 is such [2] that no change of configuration of the carboxyl substituent is likely during the conversion of gibberellic acid into epiallo- and allo-gibberic acid, where it is known to be β-situated; [2] and a formal proof of configuration at this position are advanced in the following paper.[3] Optical rotatory dispersion evidence for a β-hydrogen atom at position 10a in the methyl gibberellate hydrogenolysis product (I), and hence in gibberellic acid, has already been briefly reported [4] and will be considered in detail in a later paper.[5] This conclusion is strongly reinforced by the nuclear magnetic resonance studies described below. The 2-hydroxyl substituent in gibberellic acid is known to have the quasiaxial configuration.[6] Two possible absolute configurations, (II) and (III), for ring A must then be considered. Initially,[4] we favoured the α-oriented



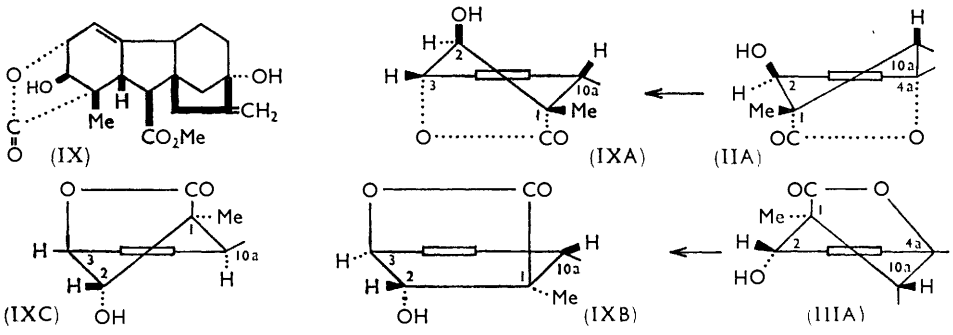
1 ⟶ 4a-lactone configuration (II) for two reasons. First, the rotatory dispersion curve (curve A, Fig. 1) for the ring A ketone (IV), obtained by oxidation of methyl tetrahydrogibberellate, showed a strong positive Cotton effect consistent with application of the octant rule to structure (IV), and similar in sign to, but of greater amplitude than, that shown by (+)-homoepicamphor [7] (V) which has an additional methyl substituent in a negative quadrant. Secondly, consideration of molecular models showed that the *trans*-A/B-fusion (II) was less strained and hence more stable than the *cis*-A/B-fusion of (III): and structure (II) was considered to account more satisfactorily for the stability of the lactone ring and its ease of reclosure in the ring A reduction products of gibberellic acid than did the alternative (III) with the 5-membered rings *trans*-fused and more rigidly locked. The α-oriented lactone (II) was also favoured by Stork and Newman [8] on the basis of their observed molecular rotation difference of +75° (lactone–acid) between the 2(*eq*)-hydroxy-epimer (VI; R = R′ = Me, R″ = H) of methyl tetrahydrogibberellate and the dimethyl ester (VII) of the dibasic acid obtained by hydrolysis of the lactone ring.

* Part XIX, *J.*, 1962, 7.

[1] Grove, MacMillan, Mulholland, and Turner, *J.*, 1960, 3049.
[2] Grove and Mulholland, *J.*, 1960, 3007.
[3] Bourn, Grove, Mulholland, Tidd, and Klyne, following paper.
[4] Cross, Grove, McCloskey, Mulholland, and Klyne, *Chem. and Ind.*, 1959, 1345.
[5] Aldridge, Grove, McCloskey, and Klyne, in the press.
[6] Cross, Grove, and Morrison, *J.*, 1961, 2498.
[7] Ourisson, quoted by Klyne, *Tetrahedron*, 1961, **13**, 29.
[8] Stork and Newman, *J. Amer. Chem. Soc.*, 1959, **81**, 5518.

This interpretation, in terms of Klyne's [9] extension of Hudson's lactone rule, was criticised by Edwards *et al.*[10] who, by analogy with the molecular rotation differences (lactone–acid) observed for certain diterpene lactones of known absolute configuration, deduced from Stork and Newman's evidence that the lactone bridge in gibberellic acid was β-oriented. The available evidence suggests that the extended Hudson rule is inapplicable to bridged lactones of the gibberellic acid type where, in the absence of a hydrogen atom at the asymmetric centre carrying the potential hydroxyl group, there is ambiguity in selecting one of the two fused carbocyclic rings as reference ring system. Nevertheless, we have failed to prepare the ester (VII) according to the directions of Stork and Newman, who worked with mixtures of 8-epimers as judged by the physical constants quoted [8] (see Experimental section).

A detailed examination of the substitution and elimination reactions of the 2-hydroxy-substituent in the ester (I) indicated [11,12] that hydrogenolysis of methyl gibberellate was accompanied by a change in the conformation of ring A. The simplest interpretation [12] of these results was based on a *trans*-fusion of rings A/B in the ester (I) and supported the β-orientation (III) of the lactone ring in gibberellic acid; but another explanation, consistent with the alternative α-orientation (II), will now be proposed.[5]



Masamune [13] used Prelog's atrolactic acid method [14] to determine the absolute configuration of the 2-hydroxy-substituent in methyl 8-epitetrahydrogibberellate and in its 2-hydroxy-epimer and he concluded that the lactone bridge was α-oriented. However, no great weight can be attached to this evidence in view of the low rotation (+3·6° and −2·2°, respectively) of the atrolactic acid, which was isolated in only 71% yield. Atrolactic acid of much greater rotation is usually obtained in those cases [14] where the L, M, and S groups (Prelog's notation) can be identified unequivocally.

The work now described establishes the α-orientation of the lactone bridge in gibberellic acid by showing that the A/B-fusion is *trans*.

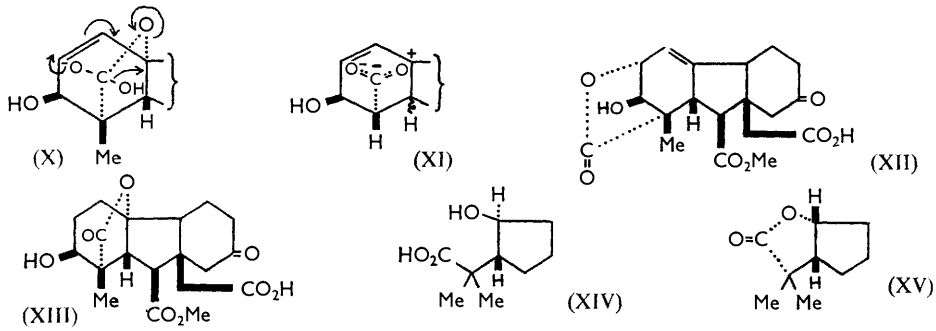
First, two aspects of the chemistry of the gibberellins are difficult to explain on the *cis*-A/B structure (III): (*a*) The ready dehydration of the 2(*ax*)-hydroxy-acid, gibberellin A_1 (VI; R = H, R′R″ = CH_2) to a gibb-2-ene, gibberellin A_5 (VIII),[15] would be unlikely since the latter would be in a strained boat conformation; (*b*) the base-catalysed rearrangement of methyl gibberellate to the gibb-4-ene-1 ⟶ 3-lactone (IX), which takes place at room temperature,[6] is readily explicable in terms of the ring A conformations (IIA) ⟶ (IXA); but the β-lactone bridge in (IXB), derived from (IIIA), would be formed from a

[9] Klyne, *Chem. and Ind.*, 1954, 1198.
[10] Edwards, Nicolson, Apsimon, and Whalley, *Chem. and Ind.*, 1960, 624.
[11] Grove, *Quart. Rev.*, 1961, **15**, 56.
[12] Cross, Grove, McCloskey, MacMillan, Moffatt, and Mulholland, " Advances in Chemistry Series," Amer. Chemical Society, Washington, 1961, Vol. XXVIII, p. 3.
[13] Masamune, *J. Amer. Chem. Soc.*, 1961, **83**, 1515.
[14] Dauben, Dickel, Jeger, and Prelog, *Helv. Chim. Acta*, 1953, **36**, 325, and earlier papers.
[15] MacMillan, Seaton, and Suter, *Tetrahedron*, 1960, **11**, 60.

carboxyl substituent which had become equatorial as a result of the rearrangement, forcing ring A into the very highly strained, and therefore unlikely, half-boat conformation (IXB). If (III) correctly represented the stereochemistry of gibberellic acid, it seemed most improbable that this reaction would take place other than by a mechanism giving simultaneous inversion of configuration at position 10a with the formation of (IXC). In order to test this hypothesis the rearrangement of methyl gibberellate to the gibb-4-ene (IX) was carried out in deuterium oxide. No evidence for the presence of a carbon–deuterium bond in the product was detected by infrared spectroscopy; and the nuclear magnetic resonance spectrum of the acetyl derivative was identical with that of undeuterated material,[16] particularly in the region of the quartet, near $\tau = 7$, due to the 10- and 10a-hydrogen atoms. This showed that there had been no incorporation of deuterium, and hence no inversion at position 10a.

This experiment provided strong evidence in favour of the absolute configuration (II). It also showed that rearrangement of gibb-3-ene-1 ⟶ 4a-lactone to gibb-4-ene-1 ⟶ 3-lactone involved only carbon atoms 3, 4, and 4a and the lactone bridge, and supported a concerted cyclic mechanism (X) for this reaction following the addition of OH^- to the lactone C=O group; but the alternative non-concerted type of allylic shift involving an ion-pair intermediate [17] of type (XI) is not wholly excluded.

A reaction mechanism involving an intermediate 4a-carbonium ion could lead to inversion at position 4b as well as at 10a, and molecular models show no great difference in stability between the 4b-epimers of structure (IX). That the configuration at 4b in the gibb-4-ene (IX) is the same as in methyl gibberellate is shown by the non-incorporation of deuterium during the rearrangement in deuterium oxide, and by the optical rotatory dispersion curve of the ring-D seco-acid (XII) [6] which showed a Cotton effect ($10^{-2}a$, $+45°$) similar in sign and amplitude to that ($10^{-2}a$, $+49°$) of the corresponding seco-compound (XIII) [18] derived from gibberellin A_1 methyl ester.



Secondly, in support of our " relactonisation " argument,[4] which was criticised by Edwards *et al.*,[10] all attempts to convert the racemic *trans*-hydroxy-acid (XIV) into a *trans*-fused lactone, analogous to that postulated in (III), failed; but heating the hydroxy-acid at 200° gave the *cis*-lactone (XV) characterised as the anilide of the corresponding *cis*-hydroxy-acid. Comparison of the rates of alkaline hydrolysis of the *cis*-lactone (XV) and the lactone system of gibberellin A_5 showed that, although the model lactone was opened appreciably faster, relactonisation occurred immediately on acidification and the corresponding hydroxy-acids were not isolated. The behaviour of the lactone bridge in gibberellin A_5 is thus in accord with the *trans*-A/B-structure (VIII).

A third piece of evidence, conclusively in favour of the absolute configuration (II),

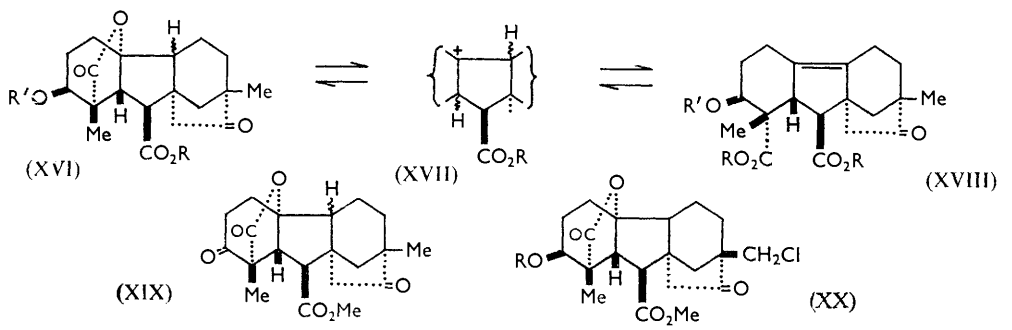
[16] Sheppard, *J.*, 1960, 3040.
[17] Sneen and Rosenberg, *J. Amer. Chem. Soc.*, 1961, **83**, 895; Winstein, Clippinger, Fainberg, Heck, and Robinson, *J. Amer. Chem. Soc.*, 1956, **78**, 328.
[18] Seta, Kitamura, Takahashi, and Sumiki, *Bull. Agric. Chem. Soc. Japan*, 1957, **21**, 73.

resulted from a reaction involving an intermediate gibbane 4a-carbonium ion. The acid-catalysed rearrangement of gibberellin A_1 has been shown [19,20] to give two isomeric lactonic keto-acids (XVI; R = R′ = H) referred to below as *A* {m. p. 265—267° (decomp.); methyl ester, m. p. 226—228°, $[\alpha]_D$ +50°} and *B* {m. p. 268—270° (decomp.); methyl ester, m. p. 183—185°, $[\alpha]_D^{18}$ +24°}. In addition to these two products, an unsaturated keto-ester, $C_{21}H_{28}O_6$ (XVIII; R = Me, R′ = H), previously obtained [19] by the action of methanolic hydrogen chloride on keto-ester *A*, is also formed (after methylation) in this reaction and is readily separated from the lactonic keto-esters by chromatography. The lactonic keto-esters could not be separated by this means, and the more soluble isomer *B* was isolated only by laborious fractional crystallisation. The composition of a mixture of these keto-esters was estimated to within ±10% by measurement of the optical rotation. The position of the double bond in the keto-ester (XVIII; R = Me, R′ = H) and the stereochemical relation of the lactonic keto-esters is apparent from the following results.

Keto-esters *A* and *B* were mutually interconvertible by treatment with dilute mineral acid. So far as could be ascertained from the crude analytical method employed, 2 hours' boiling with 2N-hydrochloric acid, followed by methylation of the product, resulted in the same equilibrium mixture of keto-esters *A* and *B* and the unsaturated ester (XVIII; R = Me, R′ = H) in the ratio 5 : 2 : 3; and no other products were detected. When the reaction time was increased to 168 hr. the same three products were obtained in approximately the same ratio; but some decomposition took place and the presence of intractable gummy material made analysis of the mixture difficult. Hydrolysis of the ester (XVIII; R = Me, R′ = H) with dilute mineral acid gave keto-acid *A* as well as non-lactonic products (XVIII; R = H or Me; R′ = H).

These results show that the 4a-carbonium ion (XVII) is an intermediate in the interconversion of the lactonic keto-esters *via* the unsaturated keto-ester (XVIII; R = Me, R′ = H), and the lactonic esters may therefore differ in configuration at position 4b or 10a or at both centres. In the conversion of gibberellin A_1 into keto-acid *A*, stereochemical changes may occur at these centres in addition to the Wagner–Meerwein rearrangement of rings C/D.

Oxidation of keto-ester *B* with chromic oxide gave a diketo-ester, $C_{20}H_{24}O_6$, m. p. 215°, isomeric with the keto-ester (XIX), m. p. 218—219°, obtained by a similar oxidation [19]



of keto-ester *A*. Subtraction of the optical rotatory dispersion curves (E and G, Fig. 2) for keto-esters *A* and *B* from the curves (D and F, Fig. 2) for the corresponding diketo-esters (XIX) gave curves B and C (Fig. 1) which were very similar in shape and amplitude to the curve (A, Fig. 1) for the ketone (IV). Keto-esters *A* and *B* do not, therefore, differ in configuration at position 10a and must be epimeric at 4b. This conclusion is confirmed by the magnitude of $J_{10,10a}$ in the proton magnetic resonance spectrum (see below) of keto-ester *B*, indicative of the same *trans*-relationship at these centres as in methyl

[19] Cross, *J.*, 1960, 3022.
[20] Takahashi, Seta, Kitamura, Kawarada, and Sumiki, *Bull. Agric. Chem. Soc. Japan*, 1957, **21**, 71.

gibberellate,[16] and by the following evidence showing that keto-ester *A* and the gibberellins also have the same configuration at position 10a.

The 8-methylene substituent of gibberellin A_1 methyl ester is readily attacked by electrophilic reagents and the formation of an 8-carbonium ion is followed by the rearrangement of rings C–D. Thus, t-butyl hypochlorite in a non-acidic medium gave the chloroketone (XX; R = H) which, since only centres 7 and 8 are involved in the reaction, has the same configuration at positions 4b and 10a as gibberellin A_1.

In addition to two three-proton singlets at τ 8·85 and 6·20, due to CMe and OMe groups, the nuclear magnetic resonance spectrum (Table 1) of the acetyl derivative (XX; R = Ac) showed a two-proton singlet at τ 6·40, ascribed to the $-CH_2Cl$ group, and a quartet of lines at τ 6·73, 6·90, 7·28, 7·43 with J = 7 c./sec., ascribed to the hydrogens at positions 10 and 10a. There was also a one-proton unresolved multiplet at τ ~7·1 and a broad peak at τ 7·68, ascribed to the hydrogen at position 4b and the β-hydrogen at position 9,

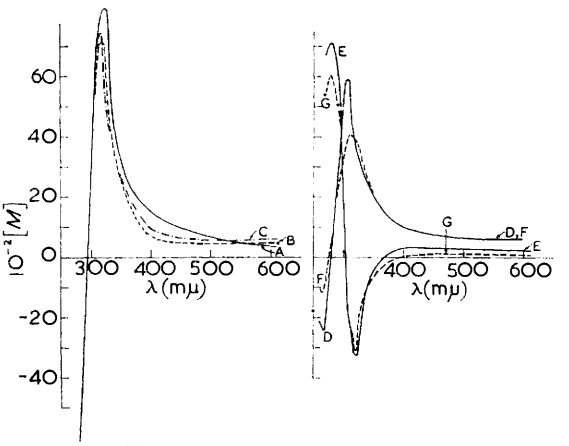


FIG. 1. FIG. 2.

FIG. 1. Optical rotatory dispersion: (A) ketone (IV); subtraction curves, (B) (– – –) and (C) (— . — . —).

FIG. 2. Optical rotatory dispersion curves for the keto-esters: (XVI; R = Me, R′ = H), curve E for A, curve G for B; and (XIX), curve D for A, curve F for B.

respectively. Except for the absence of $-CH_2Cl$ peak at τ 6·40 and the presence of a second CMe peak at τ 8·92, the spectrum of the acetyl derivative (XVI; R = Me, R′ = Ac) of keto-ester *A* was almost identical with $\tau_{10,10a}$ = 6·73, 6·88, 7·29, 7·43 ($J_{10,10a}$ = 7 c./sec.), τ_{4b} 7·20, and $\tau_{9\beta}$ 7·70. The spectrum of the acetyl derivative of keto-ester *B* was markedly different, showing $\tau_{10,10a}$ 6·55, 6·83, 7·08, 7·38 ($J_{10,10a}$ = 11 c./sec.), and a broad two-proton peak at τ 7·61, attributed to the unresolved 9β- and 4b-hydrogen resonance. The differences in τ for the hydrogen at positions 10 and 10a, and in $J_{10,10a}$ in the two isomers, arise from different ring-B conformations since the configuration at positions 10 and 10a is unaltered.

These results show that keto-ester *A* has the same configuration at 4b as the chloroketone (XX; R = H) and the gibberellins; the isomer *B* is the 4b-epimer. The reisolation in these reactions of compounds unchanged in configuration at position 10a under conditions where change would have been possible shows that they, and hence gibberellic acid, have the more stable *trans*-A/B-fusion (II).

In 4b-epimeric 8-oxo-7α-gibbane relatives containing an aromatic ring A, the configuration at position 4b had little effect on the amplitude of the rotatory dispersion curve;[1] but the plain curves of the products of reduction of the 8-keto-group were more strongly

positive for the " epi "-series with 4bβ, and on this basis absolute configurations could be assigned to 4b-epimeric pairs. The shapes of the curves (E and G) for keto-esters *A* and *B* were so similar as to make this impossible. The absolute configuration at this centre will be discussed in the following paper [3] where the 4bβ-configuration is assigned to keto-ester *A*.

Further evidence was derived from the nuclear magnetic resonance spectra of the products obtained when the equilibrium (XVI ⇌ XVII ⇌ XVIII) was established in N-deuterium chloride. The spectrum of the undeuterated keto-ester *A* was similar to that of the acetyl derivative, except that the one proton peak due to the RR'CH·OH group was seen at τ *ca.* 6·1 compared with τ 5·0, and the 1-methyl resonance appeared at lower field, at τ *ca.* 8·79, compared with τ 8·85, owing to deshielding by the adjacent

TABLE 1.

Chemical shifts (τ values) of protons in derivatives of gibberellin A_1 transformation products.

	Position of proton										
Compound	7	1	OAc	9	4b	10		10a		OMe	2
(XX; R = Ac)	6·40	8·85	7·88	7·68	7·1 *	7·43	7·28	6·90	6·73	6·20	5·00
(XVI; R = Me, R' = Ac) *A*	8·92	8·85	7·88	7·70	7·20	7·43	7·29	6·88	6·73	6·22	5·00
(XVI; R = Me, R' = Ac) *B*	8·97	8·97	7·88	7·61*	7·61*	7·38	7·08	6·83	6·55	6·27	5·03
(XVIII; R = Me, R' = Ac)	8·95	8·85	7·85	7·60	—		6·57	6·57		6·35 6·27	4·70

* Broad, unresolved.

hydroxyl group. The intensity of the CMe peak at τ 8·92 was much reduced in the deuterated specimen of this compound obtained from gibberellin A_1 methyl ester by boiling in N-deuterium chloride for 2 hr., confirming the allocation of the 1-methyl resonance to the peak at τ 8·79.

The peaks associated with the 4b-hydrogen resonance were absent from the deuterated specimens of keto-esters *A* and *B*, isolated after treatment of keto-ester *B* under the same conditions, showing that deuterium had been incorporated at this position. The specimen of the keto-ester (XVIII; R = Me, R' = H) isolated from the same equilibration was shown by infrared and nuclear magnetic resonance spectra, which were identical with the spectra of an undeuterated specimen, to have no deuterium incorporated. The spectrum (Table 1) of the acetyl derivative (XVIII; R = Me, R' = Ac) had only a single one-proton peak in the region τ 4—6 due to the RR'CH·OAc group, and it is consistent only with the presence of a tetrasubstituted ethylenic double bond which must therefore be in the (more stable) 4a(4b)-position.

Equilibration of keto-ester *A* with N-deuterium chloride for 1 week gave a deuterated keto-ester of the same melting point, whose nuclear magnetic resonance spectrum showed, by the absence of the peak at τ 7·7, that deuterium had been incorporated at position 9 in addition to 4b. The spectrum of the unsaturated keto-ester (XVIII; R = Me, R' = H) isolated from the same experiment showed *ca.* 50% diminution in intensity of the resonance (see below) ascribed to hydrogen at positions 10 and 10a in addition to the absence of a two-proton singlet at τ 7·6 attributed, in the undeuterated material, to the hydrogen atoms at position 9.

Finally, the β-configuration at position 10a and the α-orientation of the lactone bridge can also be deduced independently as follows: the magnitude (11 c./sec.) of the coupling constant for the 10- and 10a-hydrogen atoms in methyl acetylgibberellate was taken [16] to indicate a nearly coplanar arrangement of the C–H bonds. Although it has now been shown [21] that adjacent C–H bonds in systems of rigidly fused rings can give rise to coupling constants greater than those calculated by Karplus [22] as a function of the dihedral angle

[21] Williamson and Johnson, *J. Amer. Chem. Soc.*, 1961, **83**, 4623.
[22] Karplus, *J. Chem. Phys.*, 1959, **30**, 11; *J. Phys. Chem.*, 1960, **64**, 1793.

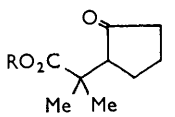
(ϕ) between them, the coupling constant is not greater than 9 c./sec. for adjacent C–H bonds with $\phi = 0°$. The magnitude of $J_{10,10a}$ in methyl acetylgibberellate and in the keto-ester *B*, therefore, indicates that these two C–H bonds are *trans*-related, a conclusion which receives further support from the following considerations.

The nuclear magnetic resonance spectrum of methyl cyclopentanecarboxylate showed a three proton singlet at τ 6·43 due to the methyl of the methoxycarbonyl group, and a one proton multiplet centred at τ 7·44 which is ascribed to the proton at position 1. The two protons at positions 2 and 5 which are *cis* to the ester group appeared as an incompletely resolved peak at τ *ca.* 8·04, on the side of the main CH_2 peak at τ 8·30. The expected τ values for the 10- and 10a-protons in the keto-esters (XVI; R = Me, R′ = H) are therefore 7·4 and ~7·6, respectively, allowing 0·7 τ unit of deshielding, compared with normal methylene hydrogen shift of 8·3 τ, for the fact that the tertiary 10a-hydrogen is at a ring junction in a rigid system.[23] The τ values found are 7·4, close to the expected value, and 6·8, respectively, the latter resonance being attributed in these compounds and in the gibberellins to the 10a-hydrogen as was the case with the ester (IX).[16] In the acetyl derivative (XVIII; R = Me, R′ = Ac) the quartet of lines seen in the keto-esters (XVI; R = Me, R′ = Ac) is replaced by a two-proton peak at τ 6·57; the 10- and 10a-protons have thus become magnetically equivalent and therefore no longer show mutual splitting of each other's resonance. The 10a-hydrogen has shifted downfield by *ca.* 0·3 τ unit owing to its becoming allylic [cf. (IX),[16] τ_{10a} 6·67] whilst the 10-hydrogen has moved downfield by *ca.* 0·8 τ unit. This shift is consistent only with a *cis*-relation of the 1-methoxycarbonyl group and 10-hydrogen in a structure with the 10a-hydrogen in the β-configuration.

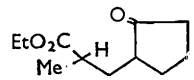
It follows that the lactone bridge must be α-oriented in the keto-esters (XVI; R = Me, R′ = H) and in gibberellic acid which has the absolute configuration (II). Thus gibberellic acid and the gibberellins, like the metabolic products of *Gibberella fujikuroi* related to (−)-kaurene,[24] has a *trans*-A/B-fusion antipodal to that more commonly present in diterpenoids.

The large deshielding, compared with the estimated τ value, of the 10a-hydrogen in the keto-esters (XVI; R = Me, R′ = Ac) is consistent with a *cis*-relation to the 10-methoxycarbonyl group; the magnitude of the effect (0·8 τ) is greater than that observed in methyl cyclopentanecarboxylate (0·3 τ) owing to the smaller separation in the rigid pentacyclic system (XVI).

The position of the RR′CH·OAc peak at τ 4·70 in the keto-ester (XVIII; R = Me, R′ = Ac) is also abnormally low and is due to the proton's being *cis* to the 1-methoxycarbonyl group, a conclusion which is in complete accord with the β-axial configuration of the 2-hydroxyl substituent in these compounds.



(XXI)



(XXII)

The model compounds (XIV) and (XV) were synthesised as follows. The keto-acid (XXI; R = H) was prepared from ethyl 2-methylhexane-2,3,6-tricarboxylate,[25] or, more directly, by alkylating the enamine from pyrrolidine and cyclopentanone with ethyl α-bromo-α-methylpropionate.[26] The enamine reaction gave two products, probably a mixture of keto-esters (XXI; R = Et) and (XXII), from which, after acid hydrolysis, the keto-acid (XXI; R = H) crystallised. Reduction of the keto-acid (XXI; R = H)

[23] Moniz and Dixon, *J. Amer. Chem. Soc.*, 1961, **83**, 1671.
[24] Cross, Galt, Hanson, and Klyne, *Tetrahedron Letters*, 1962, 145.
[25] Talukder and Bagchi, *J. Org. Chem.*, 1955, **20**, 25.
[26] Stork, Terrel, and Szmuszkovicz, *J. Amer. Chem. Soc.*, 1954, **76**, 2029; Stork and Landesman, *ibid.*, 1956, **78**, 5128.

with sodium borohydride gave a good yield of the *cis*-hydroxy-acid, which could not be isolated but was obtained as the liquid *cis*-lactone (XV) and was characterised as the anilide, m. p. 139—140°. When reduction of the keto-acid (XXI; R = H) was carried out by sodium in propan-1-ol the expected *trans*-hydroxy-acid (XIV) was obtained in high yield: with anilinomagnesium iodide it gave an anilide, m. p. 98—99°.

Experimental

M. p.s are corrected. Alumina of grade II and pH 4 was used in chromatography. Unless otherwise stated, infrared spectra were obtained for Nujol mulls and ultraviolet spectra and optical rotations for ethanol solutions. Light petroleum had b. p. 60—80°. Deuterium oxide (99·8%; d^{20} 1·10518) was supplied by Norsk Hydro. Nuclear magnetic resonance spectra were obtained for chloroform solutions (tetramethylsilane as internal standard with $\tau = 10{\cdot}00$), with Varian Associates spectrometers V-4300B (40 Mc.) or A.60 (60 Mc.).

Action of Alkali on Tetrahydrogibberellic Acid.—Tetrahydrogibberellic acid and its 8-epimer have m. p. 300—301° and 289—291°, respectively; the methyl esters of the corresponding 2(*eq*)-hydroxy-epimers, obtained by treatment with alkali,[6] have m. p. 214° and 235—239° (double m. p.), $[\alpha]_D$ +59°, and 166—168°, $[\alpha]_D$ +46°, respectively.[6,19] Stork and Newman [8] used tetrahydrogibberellic acid, m. p. 273—275°, and obtained a methyl 2(*eq*)-hydroxy ester, m. p. (185°) 196—200° $[\alpha]_D$ +34°.

Action of t-Butyl Hypochlorite on Gibberellin A_1 Methyl Ester.—Gibberellin A_1 methyl ester (200 mg.) in pyridine (2 ml.) and carbon tetrachloride (50 ml.) was treated with t-butyl hypochlorite (0·7 ml.), and the cloudy solution was set aside at 0° for 18 hr. Carbon tetrachloride (10 ml.) was added and the solution was extracted with 3N-hydrochloric acid, followed by sodium hydrogen carbonate. The recovered product was twice recrystallised from ethyl acetate–light petroleum, giving 7-*chloromethyl*-2β,4aα-*dihydroxy*-10β-*methoxycarbonyl*-1β-*methyl*-8-*oxo*-7α-*gibbane*-1α-*carboxylic acid* 1 ⟶ 4a-*lactone* (XX; R = H), prisms (90 mg.), m. p. 288—291° (Found: C, 60·9; H, 6·6; Cl, 8·8. $C_{20}H_{25}ClO_6$ requires C, 60·6; H, 6·3; Cl, 8·8%), $\nu_{max.}$ (in 1,2-dimethoxyethane) 3580, 3504 (OH); 1776, 1747, 1736 (C=O); and 741 (C–Cl) cm.$^{-1}$.

The same ester (XX; R = H) was obtained when the extraction with 3N-hydrochloric acid was omitted and water substituted: but the yield was reduced since the crude product was less clean.

The *acetyl derivative* (XX; R = Ac), prepared in acetic anhydride–pyridine at 20°, formed needles, m. p. 206—208°, from ethyl acetate–light petroleum (Found: C, 60·7; H, 6·4. $C_{22}H_{27}ClO_7$ requires C, 60·2; H, 6·2%).

Action of Hydrochloric Acid on Gibberellin A_1 Methyl Ester (cf. ref. 19).—The total product (5·9 g.) from the catalytic hydrogenation (1 mol. uptake) of methyl gibberellate (6·0 g.) was boiled with dilute hydrochloric acid for 2 hr. Methylation with diazomethane of the acidic fraction (3·90 g.) and recrystallisation of the product from ethyl acetate–light petroleum furnished the keto-ester *A* (XVI; R = Me, R′ = H) as needles, m. p. 226° (1·54 g.).

The *acetate*, prepared as above, crystallised from ethyl acetate–light petroleum in needles, m. p. 205° (Found: C, 65·6; H, 7·4. $C_{22}H_{28}O_7$ requires C, 65·3; H, 7·0%), $\nu_{max.}$ (OH absent) 1775, 1738, and 1733 cm.$^{-1}$ (C=O).

The residue (2·4 g.), on recovery, was chromatographed on alumina (30 × 3 cm.) in benzene. After intractable material (92 mg.) had been eluted with benzene (1 l.) and benzene–ether (10 : 1, 500 ml.), benzene–ether (1 : 1; 800 ml.) furnished the ester (XVIII; R = Me, R′ = H),[19] prisms (654 mg.), m. p. 157—158°, ε (mμ) 9800 (200), 8800 (205), 7500 (210), 5550 (215), 3050 (220).

The *acetate*, prepared as described above, crystallised from ether–light petroleum (b. p. 40—60°) in prisms, m. p. 132° (Found: C, 65·9; H, 7·4. $C_{23}H_{30}O_7$ requires C, 66·0; H, 7·2%), $\nu_{max.}$ (OH absent) 1739 and 1734 (sh) cm.$^{-1}$ (C=O).

Further elution of the column with ether (1 l.) followed by benzene–methanol (100 : 1; 300 ml.) afforded the keto-ester *A* (XVI; R = Me, R′ = H) (726 mg.) and, after fractional crystallisation (hand-picking) of the residues, the 4b-epimer, keto-ester *B*, prisms, m. p. 181° [19] (81 mg.). The *acetate* (XVI; R = Me, R′ = Ac), prepared as described above, crystallised from ethyl acetate–light petroleum in needles, m. p. 173—174° (Found: C, 65·4; H, 7·0. $C_{22}H_{28}O_7$ requires C, 65·3; H, 7·0%), $\nu_{max.}$ (OH absent), 1776, 1753, and 1742 cm.$^{-1}$ (C=O).

The epimeric keto-esters (XVI; R = Me, R′ = H) were stable and were recovered when chromatographed on alumina as described above, but a mixture was not separated into its components under these conditions.

Action of Hydrochloric Acid on the Keto-esters (XVI; R = Me, R′ = H).—The keto-ester (50 mg.) was heated under reflux with 2N-hydrochloric acid (4 ml.). After a given time the cooled solution was extracted with ethyl acetate and the acidic portion (usually >95%) of the product was extracted with sodium hydrogen carbonate, recovered, and methylated with diazomethane. The product was combined with the neutral fraction and recrystallised from ethyl acetate–light petroleum. After pure keto-ester *A* had been filtered off and weighed, the residue, in benzene, was chromatographed on alumina. After the unsaturated ester (XVIII; R = Me, R′ = H) had been eluted with benzene–ether (1 : 1) and weighed, a mixture of the keto-esters (XVI; R = Me, R′ = H) was obtained by elution with benzene–methanol (200 : 1) and was subjected to fractional crystallisation (hand-picking and separation of needles and prisms). As much as possible of the pure esters (XVI; R = Me, R′ = H) was obtained in this way and the composition of the residue [usually 10—15 mg. of the esters (XVI; R = Me, R′ = H) in the ratio (2 : 1)] was estimated by measurement of the optical rotation (sodium D line). The total yield of the three products was then calculated. No correction was made for losses encountered in extraction or chromatography; the crystallisation procedure led to very variable losses of material. Results are tabulated.

Keto-ester (XVI; R = Me, R′ = H)	Reaction time (hr.)	Yield (%) of products: (XVI; R = Me, R′ = H) *A*	(XVI; R = Me, R′ = H) *B*	(XVIII; R = Me, R′ = H)	Loss (%)
A	2	52	21	16	11
	168	31	10	22	37
B	2	51	20	28	1

Action of Hydrochloric Acid on the Ester (XVIII; R = Me, R′ = H).—The ester (50 mg.) in methanol (2 ml.) and 2N-hydrochloric acid (2 ml.) was boiled for 2 hr. with slow distillation of the methanol; some oil separated from the aqueous mixture. The oil was extracted with ethyl acetate, and the extract was separated into neutral [11 mg., m. p. 157°, of the ester (XVIII; R = Me, R′ = H)] and acidic (40 mg.) fractions by extraction with sodium hydrogen carbonate and recovery. Fractional crystallisation of the acidic fraction from ethyl acetate gave keto-acid *A* (XVI; R = R′ = H) (10 mg.), m. p. 255—260° (decomp.), and a fraction (4 mg.), m. p. 215—220° (decomp.), $\nu_{max.}$ 1725 cm.$^{-1}$ (broad) (no max. at 1760 cm.$^{-1}$).

Action of [^{2}H]Cl *on Gibberellin A_1 Methyl Ester*.—The ester (100 mg.) in deuterium oxide (4·5 ml.) and concentrated hydrochloric acid (0·5 ml.) was heated under reflux for 2 hr. The cooled solution was extracted with ethyl acetate saturated with deuterium oxide, and the extract was separated into neutral (12 mg. rejected) and acidic fractions by extraction with sodium hydrogen carbonate and recovery. The acidic portion (88 mg.) was methylated with diazomethane, and the product was recrystallised from ethyl acetate–light petroleum, giving needles (38 mg.), m. p. 221—223°, of 2β,4aα-dihydroxy-10β-methoxycarbonyl-1β,7-dimethyl-8-oxo[4b,7-$^2H_2$]-7α-gibbane-1α-carboxylic acid 1 ⟶ 4a-lactone, $\nu_{max.}$ 2165 cm.$^{-1}$ (C–D). The infrared spectrum was entirely different from that of the undeuterated keto-ester *A* (XVI; R = Me, R′ = H), both in the solid state and in chloroform.

Action of [^{2}H]Cl *on the Keto-esters* (XVI; R = Me, R′ = H).—(*a*) The ester *B* (100 mg.) was treated in the same way as gibberellin A_1 methyl ester (above). The acid fraction (82 mg.) was methylated; the product crystallised from ethyl acetate–light petroleum in needles (24 mg.), m. p. 224—225°, of 2β,4aα-dihydroxy-10β-methoxycarbonyl-1β,7-dimethyl-8-oxo-[4b-^{2}H]-7α-gibbane-1-α-carboxylic acid 1 ⟶ 4a-lactone, $\nu_{max.}$ 2105 cm.$^{-1}$ (C–D). The infrared spectrum, both in the solid state and in chloroform, was characteristic and differed from that of both the undeuterated keto-ester *A* (XVI; R = Me, R′ = H) and the [7,4b-$^2H_2$]-derivative.

The residue (52 mg.) was chromatographed on alumina (9 × 1 cm.) in benzene. Elution with benzene–ether (10 : 1; 150 ml.) furnished the ester (XVIII; R = Me, R′ = H), prisms (27 mg.), m. p. 156—157°. The infrared spectrum was very similar to that of undeuterated material; some small but insignificant differences were apparent in the region 8—12 μ.

Further elution of the column with benzene–methanol (400 : 1; 125 ml.; and 200 : 1, 50 ml.)

gave a gum (16 mg.) from which needles, m. p. 218—221° (2 mg.), of the above [4b-^{2}H]-keto-ester were obtained by crystallisation from ethyl acetate–light petroleum. The residue was combined with the neutral fraction (16 mg.) and chromatographed on alumina (10 × 0·7 cm.) in benzene. The fractions (30 mg.) eluted with benzene–ether (10 : 1, 300 ml.; and 1 : 1, 50 ml.) were combined and subjected to fractional crystallisation (hand-picking) from ethyl acetate–light petroleum, giving needles (rejected) and prisms (14 mg.), m. p. 174—177°, of 2β,4aα-dihydroxy-10β-methoxycarbonyl-1β,7-dimethyl-8-oxo-[4b-^{2}H]-4bα,7α-gibbane-1α-carboxylic acid 1 ⟶ 4a-lactone, $\nu_{max.}$ 2160 cm.$^{-1}$ (C–D). The infrared spectrum differed from that of the undeuterated keto-ester *B* (XVI; R = Me, R′ = H).

(*b*) The ester *A* (150 mg.) was heated under reflux for 168 hr. with deuterium oxide (9 ml.) and concentrated hydrochloric acid (1 ml.), and the mixture was worked up as before. The total extract was methylated with diazomethane, and the product (138 mg.) was crystallised from ethyl acetate–light petroleum, giving needles (97 mg.), m. p. 224—225° of 2β,4aα-dihydroxy-10β-methoxycarbonyl-1β,7-dimethyl-8-oxo-[4b,9,9-^{2}H$_3$]-7α-gibbane-1α-carboxylic acid 1 ⟶ 4a-lactone, $\nu_{max.}$ 2150 cm.$^{-1}$ (C–D). The infrared spectrum differed from that of the [4b-^{2}H]- and the [4b,7-^{2}H$_2$]-derivative. A mixed m. p. with the undeuterated ester *A* showed no depression.

The residue was chromatographed on alumina (14 × 0·7 cm.) in benzene. Elution with benzene–ether (33 : 1, 75 ml.) furnished prisms (14 mg.), m. p. 154—155°, of dimethyl 2β-hydroxy-1β,7-dimethyl-8-oxo-[9-^{2}H$_2$]-7α-gibb-4α(4b)-ene-1α,10β-dicarboxylate, $\nu_{max.}$ 2130 cm.$^{-1}$ (C–D). The infrared spectrum differed from that of the undeuterated ester (XVIII; R = Me, R′ = H).

Fractional crystallisation of the gummy product (20 mg.) eluted with benzene–ether (1 : 1, 150 ml.) gave needles (5 mg.), m. p. 222—225°, and prisms (3 mg.), m. p. 173—175°, but these materials were not further examined.

Action of NaO[^{2}H] *on Methyl Gibberellate* (cf. ref. 6).—Methyl gibberellate (100 mg.) was added to a solution of sodium hydroxide (13 mg.) in deuterium oxide (10 ml.), and the mixture was shaken for 65 min. The solution was twice extracted with ethyl acetate–ether (1 : 1) (30 ml.), and the combined organic extracts were washed with brine, dried (Na_2SO_4), and evaporated. The residue (58 mg.) crystallised from ethyl acetate–light petroleum as prisms, m. p. 173—174°. The material was recrystallised once from the same solvent system, and the infrared spectrum of the product was examined. No evidence of C–D (or O–D) bonds was found; the spectrum was indistinguishable from that of the undeuterated ester (in order to obtain evidence of O–D bonds in the product it was necessary to replace the brine in the above extraction procedure by deuterium oxide, and to use organic solvents previously washed with deuterium oxide). The product, m. p. 173—174°, was treated with pyridine (1 ml.) and acetic anhydride (200 mg.) at room temperature for 24 hr. The solvents were evaporated *in vacuo* and the residue in ether was filtered through alumina. The product crystallised from ethyl acetate–light petroleum as prisms, m. p. 166—168°.[6]

The infrared and nuclear magnetic resonance spectra showed no significant differences when compared with those of the undeuterated acetyl derivative of the ester (IX).

Oxidation of the Keto-ester B (XVI; R = Me, R′ = H) [with Dr. B. E. Cross].—The keto-ester (10 mg.) in acetone (0·5 ml.) at 20° was treated with the chromic oxide–sulphuric acid reagent [19] (0·02 ml.). After 1 hr., water was added and the product was recovered in ethyl acetate. Recrystallisation from ethyl acetate–light petroleum afforded needles (5 mg.), m. p. 215°, $[\alpha]_D^{18}$ +122° (*c* 1·0 in acetone), of 4aα-*hydroxy*-10β-*methoxycarbonyl*-1β,7-*dimethyl*-2,8-*dioxo*-4bα,7α-*gibbane*-1α-*carboxylic acid* 1 ⟶ 4a-*lactone* (XIX) (Found: C, 67·0; H, 6·9. $C_{20}H_{24}O_6$ requires C, 66·65; H, 6·7%), $\nu_{max.}$ 1771, 1742, 1733 (C=O), in $CHBr_3$ 1779 and 1730 cm.$^{-1}$. The mixed m. p. with the 4b-epimer [19] showed a large depression.

Rotatory Dispersion Curves.—Values are for [*M*] in methanol. The ketone (IV): positive Cotton effect curve; (600 mμ) +500°; (320, peak) +8200°; (265, trough) −9500°. 8-Epimer of (IV): positive Cotton effect curve; (600 mμ) +400°; (320, peak) +8250°; (265, trough) −7250°. Diketo-ester (XIX): positive Cotton effect curve; (312 mμ, peak) +4070°; (265, trough) −1330°. Diketo-ester (XIX; 4bα): positive Cotton effect curve; (307 mμ, peak) +5900° (265, trough) −2570°. Keto-ester *B* (XVI; R = Me, R′ = H): negative Cotton effect curve; (600 mμ) +100°; (320, trough) −3150°; (280, peak) +6000°; (275) +5250°. Keto-ester *A* (XVI; R = Me, R′ = H): negative Cotton effect curve; (600 mμ) +200°; (400) +300°; (320, trough) −3250°; (280, peak) +7100°; (275) +6750°.

Keto-acid (XIII): positive Cotton effect curve; (600 mμ) +100°; (310, peak,) +2600°; (267·5, trough) −2300°; (260) −2100°. Keto-acid (XII): positive Cotton effect curve; (400 mμ) +100°; (320, peak) +2400°; (277, trough) −1800°; (270, −1250°).

Gibberellin A_1: plain positive curve; (600 mμ) +200°; (400) +350°; (300) +600°. Methyl tetrahydrogibberellate: plain positive curve; (600 mμ) +50°; (400) +100°; (290) +550°.

α-Methyl-α-2-oxocyclopentylpropionic Acid.—(*a*) Dieckmann cyclisation of ethyl 2-methylhexane-2,3,6-tricarboxylate was repeated.[25] The product was not esterified but crystallised from ether–light petroleum (b. p. 40—60°) as prisms, m. p. 89—90°, of *α-methyl-α-2-oxocyclopentylpropionic acid* (XXI; R = H) (Found: C, 63·5; H, 8·3%; equiv., 160. $C_9H_{14}O_3$ requires C, 63·5; H, 8·3%; *M*, 170).

(*b*) The enamine [26] (5·2 g.) from cyclopentanone and pyrrolidine, in ethanol (50 ml.), with ethyl α-bromo-α-methylpropionate (8·2 g.) was refluxed for 16 hr. under nitrogen. Most of the ethanol was removed by distillation and the residue, with water (50 ml.), was refluxed for 1·5 hr. The mixture was extracted with ether and the product was distilled at 160°(bath)/15 mm. to give a colourless oil. The product (2·5 g.) was heated in 6N-hydrochloric acid (300 ml.) under reflux for 3 hr. The mixture was evaporated and the residual oil crystallised from ether, to give prisms (71 mg.), m. p. 89—90°, of α-methyl-α-2-oxocyclopentylpropionic acid (XXI; R = H), identified by its infrared spectrum.

Reduction of the Keto-acid (XXI; R = H).—(*a*) *Sodium in propan-1-ol.* The keto-acid (XXI; R = H) (213 mg.) in boiling propan-1-ol (15 ml.) was treated with sodium (300 mg.) during 1·5 hr. The cooled solution was acidified to pH 2 with 3N-hydrochloric acid and extracted with ethyl acetate. The product crystallised from ether–light petroleum (b. p. 40—60°) as prisms (187 mg.), m. p. 79—80°, of *α-methyl-α-trans-2-hydroxycyclopentylpropionic acid* (XIV) (Found: C, 63·0; H, 9·3%; equiv., 169. $C_9H_{16}O_3$ requires C, 62·8; H, 9·4%; *M*, 172), $\nu_{max.}$ 3375, 3315, 2635, 2575, 1690, and 1683 cm.$^{-1}$.

The *methyl ester* was an oil, b. p. 100°(bath)/0·4 mm. (Found: C, 64·1; H, 9·7; OMe, 16·8. $C_{10}H_{18}O_3$ requires C, 64·5; H, 9·7; OMe, 16·7%), ν_{ma} 3440 (OH), 1734 (C=O) cm.$^{-1}$.

Treatment of the ester with an excess of anilinomagnesium iodide in ether gave the *anilide*, of the *trans*-hydroxy-acid (XIV); this derivative crystallised from ethyl acetate–light petroleum as prisms, m. p. 98—99° (Found: C, 72·8; H, 8·7; N, 5·8. $C_{15}H_{21}NO_2$ requires C, 72·8; H, 8·6; N, 5·7%), $\nu_{max.}$ 3410 (OH), 3280, 3105, 3057, 1595, 1657, 1516, and 1304 cm.$^{-1}$.

Attempts were made to lactonise the *trans*-hydroxy-acid (XIV) as follows. (i) Treatment with an equimolar quantity of *NN'*-dicyclohexylcarbodi-imide in 1,2-dimethoxyethane at room temperature led to the formation of an anhydride (infrared spectrum). There was no evidence of lactonic material. (ii) Heating at 200° for 5 min. gave a mobile oil which was identified as the *cis*-lactone (XV) (see below) by comparison of infrared spectra. In addition, the mobile oil was converted into the anilide, m. p. 137—139°, previously obtained from the *cis*-lactone (XV). (iii) The hydroxy-acid was recovered (65%) after 3 days at room temperature in 3N-hydrochloric acid: some neutral material (22%) was obtained and was identified as the *cis*-lactone (XV).

(*b*) *Sodium borohydride.* Sodium borohydride (750 mg.) was added to a solution of the keto-acid (XXI; R = H) (250 mg.) in methanol (5 ml.). The mixture was left at room temperature for 20 hr., then diluted with water (20 ml.) and acidified with acetic acid. After extraction with ether the acidic product was separated by extraction with saturated sodium hydrogen carbonate solution. The soluble fraction was acidified with dilute hydrochloric acid and extracted with ether, which gave a mobile oil (153 mg.). Distillation of the oil at 70—80° (bath)/0·4 mm. gave *3,3-dimethyl-1-oxa-*cis*-bicyclo*[3,3,0]*octan-2-one* (XV) (105 mg.) (Found: C, 69·9; H, 9·2. $C_9H_{14}O_2$ requires C, 70·1; H, 9·1%), $\nu_{max.}$ 1766 (C=O) cm.$^{-1}$.

The *cis*-lactone (XV), after being treated with anilinomagnesium iodide (2 mol.), gave the *anilide* of the corresponding *cis*-hydroxy-acid as prisms, m. p. 139—140° (from ethyl acetate–light petroleum) (Found: C, 72·6; H, 8·6; N, 5·8. $C_{15}H_{21}NO_2$ requires C, 72·8; H, 8·6; N, 5·7%), $\nu_{max.}$ 3345 (OH), 3290, 3235, 3185, 3130, 1600, 1661, 1562, and 1311 cm.$^{-1}$.

The rate of alkaline hydrolysis of the lactone ring was measured in the following manner. The *cis*-lactone (XV) (154 mg.) in ethanol (5 ml.) and 0·1N-sodium hydroxide (20 ml., 2 mol.) was held at 25°. At intervals aliquot parts were withdrawn and titrated against standard hydrochloric acid (phenolphthalein). The hydrolysis was half-complete after 30 min. The *cis*-lactone was recovered on acidification.

Hydrolysis and Relactonisation of Gibberellin A_5.—Gibberellin A_5 [15] (82·5 mg.) was dissolved

in ethanol (1·5 ml.) and 0·1N-sodium hydroxide (7·5 ml., 3 equiv.) was added. At intervals 0·5 ml. portions were withdrawn and titrated against standard hydrochloric acid (phenolphthalein). At 25°, during 22 hr., only 0·12 equiv. of alkali was consumed by the lactone group. The temperature was then raised to 95°, and the hydrolysis was half-complete after 1·5 hr. Acidification, to pH 2, of the cooled hydrolysed material and extraction with ethyl acetate gave gibberellin A_5.

In a similar experiment with gibberellin A_1 methyl ester, the results were more complex owing to preferential hydrolysis of the ester linkage. The 2α(*eq*)-hydroxy-epimer [6] of gibberellin A_1 was recovered on acidification.

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