New Metabolites of *Gibberella fujikuroi*. Part XIX.[1] 3β,7β-Dihydroxykaurenolide

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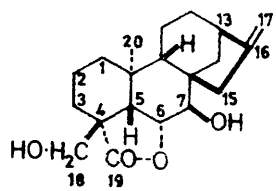
A new metabolite of *Gibberella fujikuroi* has been isolated and shown to be 3β,7β-dihydroxykaurenolide (2).

DURING the isolation of 7β,18-dihydroxykaurenolide (1)[2] by chromatography of the neutral fractions from large-scale industrial fermentations of *Gibberella fujikuroi*, an isomeric metabolite of similar polarity was discovered. A preliminary account of work leading to assignment of structure (2) to the new compound has been given.[3] †



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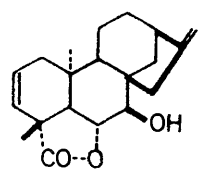

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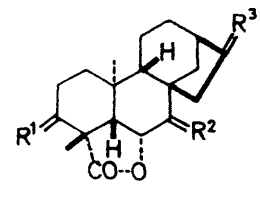


	R¹	R²	R³
(2)	α-H,β-OH	α-H,β-OH	CH_2
(3)	H_2	α-H,β-OH	CH_2
(4)	α-H,β-OH	α-H,β-OH	H,Me
(5)	α-H,β-OAc	α-H,β-OAc	CH_2
(6)	H_2	α-H,β-OH	α-H,β-Me
(7)	α-H,β-OX	α-H,β-OX	CH_2
(8)	α-H,β-OX	α-H,β-OH	CH_2
(9)	O	O	CH_2
(10)	O	O	H,Me
(11)	O	α-H,β-OH	CH_2
(12)	α-H,β-OH	O	CH_2
(13)	α-OH,β-H	α-OH,β-H	CH_2
(14)	α-OAc,β-H	α-OAc,β-H	CH_2

X = p-$MeC_6H_4·SO_2$

The i.r. spectrum of the metabolite revealed the presence of γ-lactone, terminal methylene, and hydroxy-groups [$\nu_{max.}$ ($CHBr_3$) 3610, 3470, 1763, 1662, and 886 cm^{-1}]. Its n.m.r. spectrum [τ 9·07 (3H, s, 20-H_3), 8·67 (3H, s, 18-H_3), 8·08 (d, J 7 Hz, 5-H), 5·71 (d, J 7 Hz, 7α-H), *ca.* 5·7 (m, 3-H), 5·37 (t, J 7 Hz, 6-H), and 5·14 and 5·01 (17-H_2)] closely resembled that of 7-hydroxykaurenolide (3),[4] except that it contained an additional signal, assigned to a >C*H*·OH group, and strongly suggested that the metabolite was a dihydroxykaurenolide. The presence of the terminal methylene group was confirmed by hydrogenation, which gave the dihydro-derivative (4), whose n.m.r. spectrum showed an additional methyl signal as a doublet; preparation of the diacetate (5) established the presence of the two hydroxy-groups.

The metabolite was converted into 'β-dihydro-7-hydroxykaurenolide' (6)[4] by the following reaction sequence. Treatment with toluene-p-sulphonyl chloride gave the ditoluene-p-sulphonate (7) and the monoester (8). The latter was shown to have the free

† 3β,7β-Dihydroxykaurenolide has also been isolated from fermentations of *G. fujikuroi* by Dr. J. MacMillan (personal communication).

[1] Part XVIII, B. E. Cross and R. E. Markwell, *J. Chem. Soc.* (*C*), 1971, 2980.

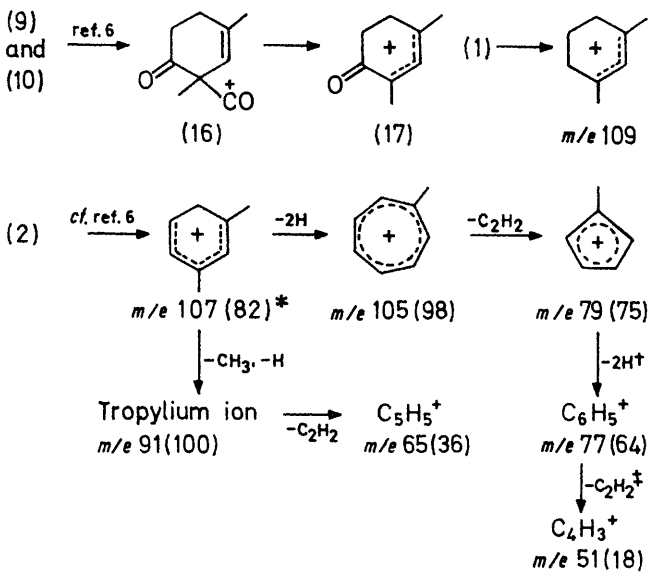
[2] B. E. Cross, J. R. Hanson, and R. H. B. Galt, *J. Chem. Soc.*, 1963, 3783.

[3] J. H. Bateson and B. E. Cross, *Tetrahedron Letters*, 1971, 3407.

[4] B. E. Cross, R. H. B. Galt, and J. R. Hanson, *J. Chem. Soc.*, 1963, 2944.

hydroxy-group at C-7 by its n.m.r. spectrum [τ 5·60 (1H, d, *J* 6·5 Hz, 7α-H) and 4·85 (t, *J* 5 Hz, 3-H)] and on treatment with boiling collidine it gave the olefin (15) [τ 4·11 (2H, m, 2-H and 3-H)]. Hydrogenation of the latter gave, after fractional crystallisation, the tetrahydro-derivative (6), identical with an authentic specimen prepared [4] from 7-hydroxykaurenolide. This inter-relationship established the carbon skeleton and stereochemistry (except for that at C-3) of the new kaurenolide.

Oxidation of the metabolite and of its dihydro-derivative (4) with chromium trioxide–pyridine–dichloromethane [5] gave the diketo-lactones (9) and (10), respectively, whereas oxidation of the metabolite with 2 equiv. of Jones reagent gave the monoketone (11) containing (n.m.r. spectrum) *ca.* 10% of the isomeric ketone (12). The mass spectra of the diketo-lactones (9) and (10) gave peaks at *m/e* 151 and 123, assigned [6] to the ions (16) and (17), thus showing that there is a keto-group in ring A of the diketo-lactones. The siting of a hydroxy-group in ring A of the dihydroxykaurenolide was confirmed by its mass spectrum (see Scheme),

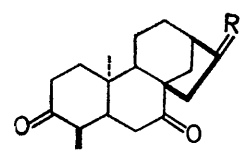


* Relative abundance in parentheses. † *m** at *m/e* 75·0. ‡ *m** at 33·8.

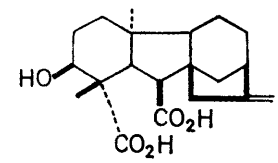
SCHEME

which was in marked contrast to that of 7-hydroxykaurenolide. The latter gave an ion at *m/e* 109 (80%) but only weak ions at *m/e* 107, 105, 91, 79, and 77.[6] The fragmentation of ring A of the dihydroxykaurenolide may be rationalised as shown in the Scheme and is supported by the presence of ions expected from the breakdown of methyltropylium and tropylium ions. Since derivatives of 3β,7β-dihydroxykaurenolide such as the Δ²-olefin (15) and the diol (13), and gibberellin A_{14} [7] (20), which has the same ring A structure, also gave strong ions at *m/e* 107, 105, 91, 79, 77, 65, and 51, this fragmentation pattern can be used to determine the structure of ring A in diterpenes and gibberellins.

The position of the ring A oxygen function was determined by reduction of the diketo-lactone (9) with chromium(II) chloride in aqueous acetone. The main product was the diketone (18), which contained a doublet in its n.m.r. spectrum at τ 9·02 (*J* 6·5 Hz) due to the 18-protons, *i.e.* decarboxylation had occurred. Consequently the oxygen function in ring A must be situated at C-3. A minor product from the reduction was the diketone (19), in which hydration of the terminal methylene group had taken place. By analogy with the hydration of kaurene,[8] the 16-hydroxy-group is provisionally assigned the α-configuration.



(18) R = CH_2
(19) R = α-OH,β-Me



(20)

The 3-hydroxy-group in the metabolite was assigned the β-configuration on the basis of n.m.r. evidence; the 5-proton doublet in its spectrum occurs at lower field (τ 8·07 in $CDCl_3$; 7·89 in pyridine) than in 7-hydroxykaurenolide (τ 8·26 in $CDCl_3$; 8·13 in pyridine), showing that the 5-proton in the former is deshielded by 1,3-diaxial interaction with a 3β-hydroxy-group.[9] In agreement, the 5-proton signal in the n.m.r. spectrum of the 3α,7α-diol (13), formed by attack by sodium borohydride on the less hindered β-face of the diketo-lactone (9) (*cf.* ref. 4), appears at τ 8·32 (in $CDCl_3$). This stereochemistry is supported by the observations that (*a*) the 3-proton in the metabolite and in its diacetate gives an n.m.r. signal at lower field (τ *ca.* 5·7 and 4·60, respectively) than in the epimeric diol (13) and its diacetate (14) (τ 6·39 and *ca.* 5·05, respectively), and (*b*) the half-band width of the 3-proton signal in the diacetate (5) is smaller ($W_{\frac{1}{2}}$ 6 Hz) than in its epimer (14) ($W_{\frac{1}{2}}$ 11 Hz); hence the 3-proton in the metabolite is equatorial and has the α-configuration.

The isolation of 3β,7β-dihydroxykaurenolide, which is the first example of a metabolite of *G. fujikuroi* in which 3-hydroxylation of a kaurenoid skeleton has occurred, suggests that there may be an alternative biosynthetic route to the gibberellins in which 3-hydroxylation precedes ring contraction.[10] This possibility is under investigation.

[5] R. Ratcliffe and R. Rodehourst, *J. Org. Chem.*, 1970, **35**, 4000.
[6] A. I. Kalinovsky, E. P. Serebryakov, A. V. Simolin, V. F. Kucherov, and O. S. Chizhov, *Org. Mass Spectrometry*, 1971, **5**, 33.
[7] B. E. Cross, *J. Chem. Soc.* (*C*), 1966, 501.
[8] J. R. Hanson, *J. Chem. Soc.*, 1963, 5061.
[9] N. S. Bhacca and D. H. Williams, 'Application of NMR Spectroscopy in Organic Chemistry,' Holden-Day, San Francisco, 1964, 183.
[10] B. E. Cross, *Progr. Phytochem.*, 1968, **1**, 195.

EXPERIMENTAL

Details of chromatographic materials and conditions used for determination of physical data, *etc.*, are reported in ref. 1.

Isolation of 3β,7β-*Dihydroxykaurenolide* (2).—Part of the crude neutral fraction (*ca.* 1300 g) from an industrial fermentation of *G. fujikuroi* was chromatographed on alumina (Laporte type O) (4 kg). Light petroleum (b.p. 40—60°; 8 l) eluted polypropylene glycol antifoam (*ca.* 800 g). Elution with ethyl acetate (*ca.* 12 l) and ethanol–ethyl acetate (1 : 4) (2 l) afforded more antifoam followed by fractions containing 7-hydroxykaurenolide [4] and cyclonerodiol.[11] Ethanol–ethyl acetate (1 : 4 and 3 : 7) eluted fractions (*ca.* 45 g) containing 7,18-dihydroxykaurenolide [2] (1) and 3β,7β-dihydroxykaurenolide (2), which were combined and rechromatographed on alumina (Laporte type O) (1700 g). Elution with ethyl acetate–light petroleum (1 : 1; 4 l) gave fractions containing antifoam and the kaurenolide (2), which after two crystallisations from ethyl acetate–light petroleum afforded 3β,7β-*dihydroxykaurenolide* (4·9 g). Recrystallisation from ethyl acetate–light petroleum gave felted needles, m.p. 175—176°, $[\alpha]_D^{24}$ −34·7° (*c* 0·64 in $CHCl_3$) (Found: C, 72·0; H, 8·4. $C_{20}H_{28}O_4$ requires C, 72·2; H, 8·5%), $\nu_{max.}$ 3490, 3280, 1736, 1660, and 895 cm^{-1}; τ (pyridine) 9·02 (3H, s, 20-H_3), 8·40 (3H, s, 18-H_3), 7·88 (1H, d, *J* 7 Hz, 5-H), 7·41 (1H, m, $W_{\frac{1}{2}}$ 8 Hz, 13-H), 6·92 (1H, dt, *J* 15 and 3 Hz, 15α-H), 5·45 (t, *J* 7 Hz, 3α-H), 5·46 (d, *J* 7 Hz, 7α-H), 5·01 (t, *J* 7 Hz, 6-H), and 4·95 (m, 17-H_2); *m/e* 332 (M^+, 28%), 314 (34), 296 (16), 121 (60), 119 (64), 107 (82), 105 (98), and 91 (100).

3β,7β-Dihydroxykaurenolide had R_F 1·1 (relative to 7,18-dihydroxykaurenolide, R_F 1·0) on t.l.c. in benzene–ethanol (9 : 1). Its *diacetate* (5), prepared with acetic anhydride in pyridine, crystallised from ethyl acetate–light petroleum in felted needles, m.p. 161—162°, $[\alpha]_D^{24.5}$ −19·6° (*c* 0·38 in $CHCl_3$) (Found: C, 69·4; H, 7·7%; *m/e* 416. $C_{24}H_{32}O_6$ requires C, 69·2; H, 7·7%; *M*, 416), $\nu_{max.}$ 1782, 1751, 1665, and 879 cm^{-1}; τ (90 MHz) 8·95 (3H, s, 20-H_3), 8·68 (3H, s, 18-H_3), 7·95 (3H, s, OAc), 7·93 (3H, s, OAc), 5·31 (1H, t, *J* 6·5 Hz, 6-H), 5·12br (1H) and 5·0br (1H) (17-H_2), 4·60 (1H, t, *J* 6 Hz, 3α-H), and 4·20 (1H, d, *J* 7 Hz, 7α-H).

Elution of the column with ethanol–ethyl acetate (1 : 19) gave fractions containing 7,18-dihydroxykaurenolide.

Gibberellin A_{14}.—Gibberellin A_{14} [8] showed *m/e* 330 (M^+, 25%), 284 (38), 269 (50), 107 (40), 105 (52), 91 (85), 79 (68), 77 (62), 65 (38), and 51 (20).

Hydrogenation of 3β,7β-*Dihydroxykaurenolide*.—The kaurenolide (105 mg) in ethyl acetate (25 ml) was reduced with hydrogen at room temperature in the presence of 5% palladium–charcoal (75 mg) until uptake ceased. The *dihydrokaurenolide* (4) crystallised from ethyl acetate–light petroleum in felted needles, m.p. 227—229° (Found: C, 71·65; H, 8·9. $C_{20}H_{30}O_4$ requires C, 71·8; H, 9·0%), $\nu_{max.}$ 3480, 3260br, and 1735 cm^{-1}; τ (90 MHz) 9·08 (3H, s, 20-H_3), 9·00 (3H, d, *J* 6·5 Hz, 17-H_3), 8·68 (3H, s, 18-H_3), 8·06 (1H, d, *J* 7 Hz, 5-H), 5·73 (1H, d, *J* 7 Hz, 7α-H), *ca.* 5·7 (1H, m, 3α-H), and 5·39 (1H, t, *J* 7 Hz, 6-H) [irradiation at the frequency of the signal at either τ 8·06 or 5·73 reduced the triplet at τ 5·39 to a doublet (*J* 7 Hz)]; *m/e* 334 (M^+, 22%), 316 (72), 298 (19), 107 (42), 105 (31), 91 (34), 79 (36), 77 (25), and 65 (13).

Reaction of 3β,7β-*Dihydroxykaurenolide with Toluene*-p-*sulphonyl Chloride*.—The kaurenolide (400 mg) was treated with toluene-*p*-sulphonyl chloride (600 mg) in pyridine (15 ml) for 10 days. Recovery in ethyl acetate gave a gum, which was chromatographed on silica gel (30 × 2·5 cm). Elution with ethyl acetate–light petroleum (5 : 35) afforded the ditoluene-*p*-sulphonate (7) as an intractable foam, $\nu_{max.}$ (film) 1770, 1665, 1600, 1500, 1370, 1190, and 860 cm^{-1} (no OH band); τ 9·13 (3H, s, 20-H_3), 8·90 (3H, s, 18-H_3), 8·03 (1H, d, *J* 6 Hz, 5-H), 7·53 (6H, s, 2 × ArMe), 5·38 (1H, t, *J* 6 Hz, 6-H), 5·03 (m) and 4·90 (m) (17-H_2), 4·93 (m, 3α-H), and 4·55 (1H, d, *J* 6 Hz, 7α-H); τ (benzene) 9·36 (3H, s, 20-H_3), 9·11 (3H, s, 18-H_3), 8·47 (d, *J* 6 Hz, 5-H), 8·01 (s, ArMe), 8·05 (s, ArMe), 5·60 (1H, t, *J* 6 Hz, 6-H), 4·97 (m) and 4·88 (m) (17-H_2), 4·71 (1H, t, *J* 5 Hz, 3α-H), and 4·40 (1H, d, *J* 6 Hz, 7α-H). Elution with ethyl acetate–light petroleum (3 : 17) gave the monotoluene-*p*-sulphonate (8), as an intractable foam (Found: *m/e* 486. $C_{27}H_{34}O_6S$ requires *M*, 486), $\nu_{max.}$ (film) 3520br, 1761, 1660, 1603, 1477, 1290, and 888 cm^{-1}; τ 9·13 (3H, s, 20-H_3), 8·76 (3H, s, 18-H_3), 8·04 (d, *J* 6 Hz, 5-H), 7·51 (3H, s, ArMe), 5·60 (1H, d, *J* 6 Hz, 7α-H), 5·32 (1H, t, *J* 6 Hz, 6-H), 5·07 (m) and 4·9 (m) (17-H_2), and 4·85 (t, *J* 5·5 Hz, 3α-H), *m/e* 107 (66%), 105 (33), 91 (100), 79 (47), 77 (35), and 65 (47).

Elimination of Toluene-p-*sulphonic Acid*.—The toluene-*p*-sulphonate (8) (304 mg) was heated under reflux in dry collidine (30 ml) for 6 h in an atmosphere of nitrogen. The solution was poured into dilute hydrochloric acid and the product was recovered in ethyl acetate and was chromatographed on silica gel (13 × 2 cm). Elution with ethyl acetate–light petroleum (1 : 9) followed by crystallisation from ethyl acetate–light petroleum gave 6α,7β-*dihydroxykaura*-2,16-*dien*-19-*oic acid* 19 → 6α-*lactone* (15) (93 mg) as prisms, m.p. 172—173° (Found: C, 76·4; H, 8·25. $C_{20}H_{26}O_3$ requires C, 76·4; H, 8·3%), $\nu_{max.}$ 3495, 1761, 1647, 896, and 719 cm^{-1} (*cis*-CH=CH−); τ 9·12 (3H, s, 20-H_3), 8·61 (3H, s, 18-H_3), 7·98 (d, *J* 7 Hz, 5-H), 5·52 (1H, d, *J* 7 Hz, 7α-H), 5·23 (1H, t, *J* 7 Hz, 6-H), 5·02 (m) and 4·89 (m) (17-H_2), and 4·11 (2H, m, $W_{\frac{1}{2}}$ 4 Hz, 2-H and 3-H); *m/e* 314 (M^+, 7%), 296 (57), 268 (60), 107 (61), 105 (92), 91 (100), 79 (73), 77 (51), 65 (26), and 51 (13).

Hydrogenation of the Δ^2-*Olefin* (15).—The Δ^2-olefin (62 mg) and 30% palladium–charcoal (60 mg) in ethyl acetate (45 ml) were shaken in hydrogen until uptake ceased. Four recrystallisations of the product from ethyl acetate–light petroleum afforded needles (13 mg), m.p. 225—227°, $[\alpha]_D^{22}$ −7·5 ± 0·7°: (*c* 0·44 in $CHCl_3$); τ 9·14 (3H, s, 20-H_3), 8·97 (3H, d, *J* 6·5 Hz, 17-H_3), 8·70 (3H, s, 18-H_3), 8·21 (d, *J* 6·5 Hz, 5-H), 5·57 (d, *J* 6·5 Hz, 7α-H), and 5·31 (t, *J* 6·5 Hz, 6-H), identical (i.r. and mass spectra, mixed m.p., and $[\alpha]_D$) with a specimen [4] of ' β-dihydro-7-hydroxykaurenolide ' (6).

Oxidation of 3β,7β-*Dihydroxykaurenolide*.—Chromium trioxide (1·8 g) was added with stirring to pyridine (3·0 ml) and dichloromethane (45 ml).[5]. Then the kaurenolide (198 mg) in dichloromethane (10 ml) was added to the solution, and stirring was continued for 15 min. The mixture was decanted and the residue was extracted with ethyl acetate. The combined solutions were washed with dilute sodium hydroxide solution, dilute hydrochloric acid, and sodium chloride solution. Recovery gave a solid which crystallised from acetone–light petroleum as needles (176 mg) of 6α-*hydroxy*-3,7-*dioxokaur*-16-*en*-19-*oic acid* 19 → 6α-*lactone* (9), m.p. 310—312° (decomp.), $[\alpha]_D^{22}$

[11] B. E. Cross, R. E. Markwell, and J. C. Stewart, *Tetrahedron*, 1971, **27**, 1663.

59·7° (*c* 0·39 in $CHCl_3$) (Found: C, 73·15; H, 7·4. $C_{20}H_{24}O_4$ requires C, 73·1; H, 7·4%), $\nu_{max.}$ 1784, 1704, 1655, and 874 cm^{-1}; τ (90 MHz) 9·24 (3H, s, 20-H_3), 8·44 (3H, s, 18-H_3), 7·22 (d, *J* 7 Hz, 5-H), 5·14 (d, *J* 7 Hz, 6-H), and 5·10 (m) and 4·93 (m) (17-H_2); *m/e* 328 (100%), 179 (13), 151 (52), and 123 (27).

Oxidation of the Dihydro-diol (4).—Treatment of the diol (4) (75 mg) with chromium trioxide (0·9 g) and pyridine (1·5 ml) in dichloromethane (22·5 ml), as above, followed by crystallisation of the product from ethyl acetate–light petroleum gave 6α-*hydroxy*-3,7-*dioxokauran*-19-*oic acid* 19 → 6α-*lactone* (10) as needles (63 mg), m.p. 313° (decomp.) (Found: C, 72·55; H, 7·7. $C_{20}H_{26}O_4$ requires C, 72·7; H, 7·9%), $\nu_{max.}$ 1772 and 1700 cm^{-1}; τ (pyridine; 90 MHz) 9·30 (3H, s, 20-H_3), 9·11 (d, *J* 6·5 Hz, 17-H_3), 8·34 (3H, s, 18-H_3), and 6·92 (1H, d, *J* 7·5 Hz, 6-H); *m/e* 330 (78%), 179 (100), 151 (94), and 123 (30).

Reduction of the Diketo-lactone (9) *with Chromium*(II) *Chloride*.—The diketo-lactone (115 mg) in acetone (60 ml) was treated with a solution of chromium(II) chloride [prepared from chromium(III) chloride hexahydrate (5 g) and zinc amalgam in dilute hydrochloric acid] in an atmosphere of nitrogen for 48 h. Removal of the acetone *in vacuo* and addition of water followed by recovery in ethyl acetate gave a gum (79 mg), which was chromatographed on silica gel (12 × 1·5 cm). Elution with ethyl acetate–light petroleum (1 : 7) followed by crystallisation from light petroleum gave 3,7-*dioxo*-19-*norkaur*-16-*ene* (18) (33 mg) as needles, m.p. 88—90° (Found: C, 79·7; H, 8·9%; *m/e* 286. $C_{19}H_{26}O_2$ requires C, 79·7; H, 9·1%; *M*, 286), $\nu_{max.}$ ($CHBr_3$) 1705, 1660, and 884 cm^{-1}; τ 9·02 (3H, d, *J* 6·5 Hz, 18-H_3), 8·71 (3H, s, 20-H_3), and 5·06 (2H, $W_{\frac{1}{2}}$ 7 Hz, 17-H_2).

Further elution of the column, with ethyl acetate–light petroleum (1 : 1), followed by crystallisation from ethyl acetate–light petroleum gave 3,7-*dioxo*-19-*norkauran*-16α-*ol* (19) as prisms (28 mg), m.p. 90—92·5° (decomp.) (Found: C, 74·8; H, 9·25%; *m/e* 304. $C_{19}H_{28}O_3$ requires C, 75·0; H, 9·3%; *M*, 304), $\nu_{max.}$ 3260br, 1712, and 1697 cm^{-1}. A polymorph, m.p. 94—98° (decomp.), showed $\nu_{max.}$ 3340br, 1701, and 1695sh cm^{-1}; τ (90 MHz) 9·00 (3H, d, *J* 6 Hz, 18-H_3), 8·60 (s) and 8·62 (s) (6H, 17-H_3 and 20-H_3), and 7·29 (1H, d, *J* 14 Hz, 15α-H).

Oxidation of the Dihydroxykaurenolide (2) *with Two Equivalents of Jones Reagent*.—The kaurenolide (97 mg) in acetone (25 ml) was treated with the 8N-chromium trioxide reagent (79 μl; 2·2 equiv.) at 0° for 30 min. The identical (i.r. spectrum) with the sample prepared before. Further elution gave material which crystallised from ethyl acetate–light petroleum as rosettes of needles (35 mg), which on recrystallisation gave 6α,7β-dihydroxy-3-oxokaur-16-en-19-oic acid 19 → 6α-lactone (11), containing (n.m.r. spectrum) *ca.* 10% of the 3β-hydroxy-7-oxo-isomer (12), as needles, m.p. 205—208° (Found: C, 72·3; H, 7·8%; *m/e* 330. Calc. for $C_{20}H_{26}O_4$: C, 72·7; H, 7·9%; *M*, 330), $\nu_{max.}$ ($CHCl_3$) 3450, 1759, 1700, 1650, and 890 cm^{-1}; τ (90 MHz) 9·12 (3H, s, 20-H_3), 8·53 (3H, s, 18-H_3), 7·69 (1H, d, *J* 6·5 Hz, 5-H), 5·62 (1H, d, *J* 6·5 Hz, 7α-H), 5·43 (1H, t, *J* 6·5 Hz, 6-H), and 5·13br (1H) and 4·97br (1H) (17-H_2); the 3β-hydroxy-7-oxo-isomer showed τ 9·22 (s, 20-H_3) and 8·60 (s, 18-H_3).

Further elution of the column gave the kaurenolide (2), m.p. 174—176°.

Reduction of the Diketo-lactone (9) *with Sodium Borohydride*.—Sodium borohydride (400 mg) was added in portions, with stirring, to the diketo-lactone (110 mg) in methanol (35 ml) at 0° and the solution was left at room temperature overnight. 2N-Acetic acid (10 ml) was added and the methanol removed *in vacuo*. Recovery in ethyl acetate, followed by crystallisation from ethyl acetate–light petroleum, gave 3α,6α,7α-*trihydroxykaur*-16-*en*-19-*oic acid* 19 → 6α-*lactone* (13) as prisms (95 mg), m.p. 230—232° (Found: C, 71·9; H, 8·4%; *m/e* 332. $C_{20}H_{28}O_4$ requires C, 72·2; H, 8·5%; *M*, 332), $\nu_{max.}$ 3440, 1742, 1659, and 879 cm^{-1}; τ (90 MHz) 8·82 (3H, s, 20-H_3), 8·48 (3H, s, 18-H_3), 8·32 (d, *J* 5 Hz, 5-H), 7·38 (1H, m, $W_{\frac{1}{2}}$ 8 Hz, 13-H), 6·39 (1H, q, $W_{\frac{1}{2}}$ 11 Hz, 3β-H), 5·85 (1H, d, *J* 8 Hz, 7β-H), 5·0 and 5·15 (17-H_2), and 5·05 (q, *J* 5 and 8 Hz, 6-H); *m/e* 332 (M^+, 32%), 314 (38), 296 (25), 172 (75), 171 (95), 107 (81), 105 (65), 93 (80), 91 (100), 79 (85), 77 (64), 65 (31), and 51 (10).

Its *diacetate* (14), obtained by treatment with acetic anhydride in pyridine for 15 days, crystallised from ethyl acetate–light petroleum as prisms, m.p. 155—157° (Found: C, 69·0; H, 7·7. $C_{24}H_{32}O_6$ requires C, 69·2; H, 7·7%), $\nu_{max.}$ 1780, 1760, 1723, 1657, and 890 cm^{-1}; τ 8·83 (3H, s, 20-H_3), 8·61 (3H, s, 18-H_3), 8·27 (1H, d, *J* 5 Hz, 5-H), 7·86 (3H, s, OAc), 7·83 (3H, s, OAc), 7·36 (1H, m, $W_{\frac{1}{2}}$ 8 Hz, 13-H), 5·06 (q, *J* 5 and 8·5 Hz, 6-H), *ca.* 5·05 (m, 3β-H), 5·07br and 4·95br (17-H_2), and 4·72 (1H, d, *J* 8·5 Hz, 7β-H).

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