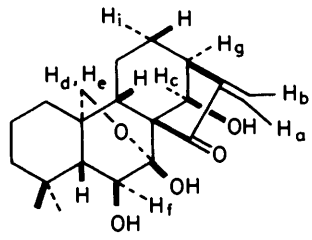
Structure Elucidation of Longikaurin A and Longikaurin B, New Biologically Active Diterpenoids from *Rabdosia longituba*[1] and Chemical Conversion of Oridonin into Dihydrolongikaurin A

Yoshio Takeda * and Tetsuro Fujita
Faculty of Pharmaceutical Sciences, The University of Tokushima, Tokushima 770, Japan
Tetsuro Shingu
Faculty of Pharmaceutical Sciences, Kobe-Gakuin University, Nishi-ku, Kobe 673, Japan

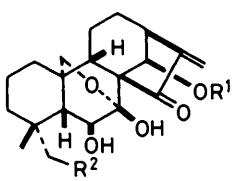
Biologically active diterpenoids longikaurin A (**1**) and longikaurin B (**2**) were isolated, together with the known kamebakaurin (**5**), from the leaves of *Rabdosia longituba* and their structures were elucidated from spectral and chemical evidence. Oridonin (**9**) was chemically converted into (16*R*)-dihydrolongikaurin A (**16**).

The leaves of *Rabdosia longituba* (Miquel) Hara[2] are intensely bitter. The bitter principles, nodosin (**6**), isodocarpin (**7**), lasiokaurin (**8**), and oridonin (**9**), have already been isolated.[3] In the course of our studies on biologically active substances from plants belonging to the genus *Rabdosia* (Labiatae), we examined the constituents of the leaves of *R. longituba* collected in Tokushima Prefecture (Japan), and isolated two new compounds, longikaurin A (**1**) and longikaurin B (**2**), along with the known kamebakaurin (**5**),[4] as major diterpenoids. Longikaurin A (**1**) showed cytotoxic activity against cultured rat mammary cancer cells FM 3A/B, and both longikaurin A (**1**) and longikaurin B (**2**) showed antibacterial activity against gram-positive bacteria.[5] This report deals with structure elucidation of the two new diterpenoids and chemical conversion of oridonin (**9**) into (16*R*)-dihydrolongikaurin A (**16**).

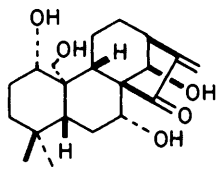
Longikaurin A (**1**), $[\alpha]_D$ −91.1° (C_5H_5N), was isolated as needles, m.p. 223—225 °C, and the molecular formula was determined as $C_{20}H_{28}O_5$ from the results of elemental analysis and mass spectra [electron impact and chemical ionization (EI and CI)]. Longikaurin A (**1**) was presumed to contain a five-membered-ring ketone conjugated with an exocyclic methylene group as a partial structure from the following spectral data: u.v. [$\lambda_{max.}$(MeOH) 236 nm], i.r. [$\nu_{max.}$(Nujol) 1 705 and 1 640 cm^{-1}], ^1H n.m.r. [δ($CDCl_3$) 5.52 (H_b) and 6.14 (H_a) (each 1 H, br s)], and ^{13}C n.m.r. [δ(C_5D_5N) 120.1 (t), 153.5 (s), and 209.3 (s)] (see Table 1). The ^1H n.m.r. spectrum of longikaurin A (**1**) showed



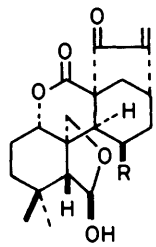
(**1**)



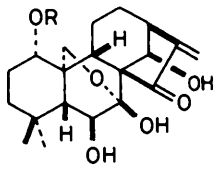
(**1**) $R^1 = R^2 = H$
(**2**) $R^1 = H, R^2 = OAc$
(**3**) $R^1 = Ac, R^2 = H$
(**4**) $R^1 = R^2 = Ac$



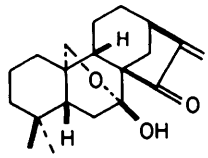
(**5**)



(**6**) R = OH
(**7**) R = H



(**8**) R = Ac
(**9**) R = H



(**10**)

Table 1. ^{13}C N.m.r. data[a] for longikaurins A (**1**) and B (**2**)

Carbon	(1)	(2)
1	31.3[b]	30.9[c]
2	17.3	17.4
3	42.1	36.5
4	34.5	37.1
5	53.1	52.8
6	74.8	73.8
7	98.9	99.0
8	63.2	63.1
9	61.4	62.0
10	37.1	38.1
11	19.6	19.2
12	30.8[b]	30.6[c]
13	44.4	44.4
14	74.1	74.1
15	209.3	209.3
16	153.5	153.3
17	120.1	120.2
18	34.2	28.0
19	23.0	67.4[d]
20	66.8	67.2[d]
CH_3CO		21.3; 171.4

[a] Measured for C_5D_5N solution; [b–d] Assignments may be interchanged.

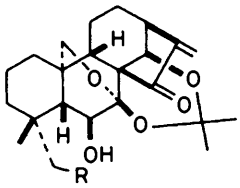
Table 2. Results of INDOR spectra and n.O.e. experiments for longikaurin A (**1**) (measured for $CDCl_3$ solution)

Monitor proton	Protons which showed n.O.e. for monitoring proton (%)	Protons which showed coupling with monitored proton
H_a	H_b (32)	H_b, H_g
H_b	H_a (31)	H_a, H_g
H_c		H_g
H_d	Me_2 (δ 1.09)	H_e
H_f[a]	Me_2	
H_g		H_b, H_c, H_i
H_i		H_g
Me_2	H_f (6.4)	

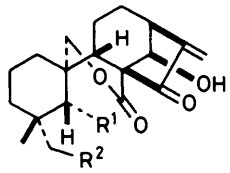
[a] Monitored after D_2O treatment.

the presence of three hydroxy groups [δ 4.90 (1 H, m), 6.10 (1 H, m), and 6.59 (1 H, d, *J* 10 Hz), disappeared on addition of D_2O]. The ^1H n.m.r. [δ 3.77 (1 H, d, *J* 7 Hz, after D_2O exchange, H_f) and 4.76 (1 H, d, *J* 2 Hz, H_C)] and ^{13}C n.m.r. [δ 74.1 and 74.8 (each doublet)] spectra also showed the presence of two secondary hydroxy groups in longikaurin A (**1**). This fact suggests that, of the three hydroxy groups in compound (**1**), two are secondary and one is tertiary. The nature of four of the five oxygen atoms thus became clear. Another oxygen was assumed to be present as an ether linkage and this assumption was supported from the fact that signals due to a $-CH_2O-$ group were observed in the ^1H n.m.r. [δ 3.81 (H_d) and 4.07 (H_e) (each 1 H, AB d, *J* 10 Hz)] and ^{13}C n.m.r. [δ 66.8 (t)] n.m.r. spectra of (**1**). Since the nature of all oxygen atoms had now been elucidated, it was deduced that longikaurin A has a pentacyclic ring system. The ^{13}C n.m.r. spectrum further showed a signal due to a ketone acetal carbon (δ 98.9). Considering the structures of many diterpenoids from *Rabdosia* species,[6] longikaurin A (**1**) was presumed to have, as a basic skeleton, *ent*-7β,20-epoxy-7α-hydroxykaur-16-en-15-one (**10**), and a structure in which two secondary hydroxy groups are added to the basic skeleton (**10**). In order to verify this presumption and to determine the positions of the two hydroxy groups, internuclear double-resonance (INDOR) experiments[7] and some chemical reactions were performed on longikaurin A (**1**). The results of INDOR experiments were shown in Table 2. When monitored on the frequencies of H_a, H_b, and H_g [δ_H 3.03 (1 H, br d, *J* 10 Hz)], INDOR signals were observed for H_b and H_g, H_a and H_g, and H_b, H_c, and H_i [δ_H 2.50 (1 H, m)] respectively. This result not only confirmed the structure of a five-membered-ring ketone conjugated with an exocyclic methylene group, but also suggested the presence of a secondary carbinyl proton, H_c, at C-14. Acetylation of longikaurin A (**1**) with acetic anhydride and pyridine gave the monoacetate (**3**), m.p. 233—235 °C, the ^1H n.m.r. spectrum of which showed the signal due to H_c at δ 5.68 (1 H, d, *J* 1 Hz), resonating at an abnormally low field compared with those for protons attached to a carbon bearing an acetoxy group. This fact showed the presence of H_c at C-14$_\alpha$ where H_c is subjected to an anisotropic effect by the carbonyl group at C-15 and has a dihedral angle of *ca.* 90° with 13-H. Hence we postulated the presence of a secondary hydroxy group at C-14$_\beta$. Treatment of longikaurin A (**1**) with 2,2-dimethoxypropane in *N*,*N*-dimethylformamide (DMF) in the presence of a catalytic amount of toluene-*p*-sulphonic acid (PTSA) gave an acetonide (**11**), m.p. 194—196 °C, thus confirming chemically the presence of a β-oriented hydroxy group at C-7 in longikaurin A. In the INDOR experiments, a signal due to nuclear Overhauser enhancement (n.O.e.) for tertiary methyl groups (δ 1.09, 6 H) was observed when monitored on the frequency of H_f. On irradiation of the signal at δ 1.09, an n.O.e. (6.4%) was observed for H_f. These results, together with the coupling pattern of H_f, allowed us to assign H_f to a proton at C-6$_\alpha$. Oxidation of longikaurin A (**1**) with sodium metaperiodate gave an aldehyde lactone (**13**), m.p. 145—147 °C and a carboxylic acid lactone (**14**), m.p. 195—197 °C, confirming the presence of a *cis*-glycol system at C-6 and C-7 in compound (**1**). Catalytic hydrogenation of longikaurin A (**1**) gave two dihydro compounds, (**16**) [δ_H 1.11 (3 H, d, *J* 9 Hz)] and (**17**) [δ_H 1.32 (3 H, d, *J* 8 Hz)]. The configurational assignments of the methyl group at C-16 were achieved by examining a stereomodel. If the secondary methyl group has an α-orientation, the ^1H n.m.r. signal is expected to be observed at a lower field than that of a β-oriented methyl due to an anisotropic effect by the ketone at C-15. Accordingly, compound (**16**), showing the signal of the newly formed secondary methyl group at higher field, was assigned 16*R* stereochemistry and compound (**17**), which exhibited the signal at lower field, was assigned as the 16*S* epimer. Both dihydro compounds, (**16**) and (**17**), showed a negative Cotton effect in the o.r.d. spectrum, which showed that the absolute configuration of the D-ring is β. Accordingly, longikaurin A has the structure *ent*-7β,20-epoxy-6α,7α,14α-trihydroxykaur-16-en-15-one (**1**).

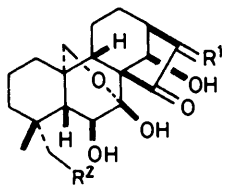
Since the structure of longikaurin A (**1**) corresponds to the 1-deoxy derivative of oridonin (**9**), we tried to correlate oridonin (**9**) with longikaurin A (**1**) chemically. *ent*-7β,20-Epoxy-6α,7α,14α-trihydroxykaurane-1,15-dione (**19**),[8] easily obtained from oridonin (**9**), was treated with ethane-1,2-dithiol and boron trifluoride–diethyl ether to give a mono(dithioacetal) (**20**). The site of dithioacetalization was determined by comparing the o.r.d. curve of the starting material (**19**) with that of the product (**20**). Thus, compound (**19**), having a ketone group at C-1, showed a positive Cotton effect at 324 nm and a negative Cotton effect at 318 nm, where as compound (**20**) showed only a negative Cotton effect at 318 nm in the o.r.d. spectrum. These results showed that only the ketone at C-1 suffered dithioacetalization, and that the ketone at C-15 remained intact. The 1-dithioacetal (**20**) thus obtained was then subjected to a desulphurization reaction by Raney-nickel to give two products, of which the more polar compound was identical with (16*R*)-dihydrolongikaurin A (**16**) on comparison of i.r. and ^1H n.m.r. spectra. Thus, the structure of longikaurin A



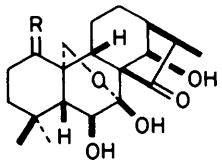
(11) R = H
(12) R = OAc



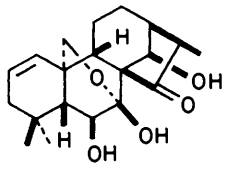
(13) R^1 = CHO, R^2 = H
(14) R^1 = CO_2H, R^2 = H
(15) R^1 = CHO, R^2 = OAc



(16) R^1 = α-H, β-Me, R^2 = H
(17) R^1 = β-H, α-Me, R^2 = H
(18) R^1 = α-H, β-Me, R^2 = OAc

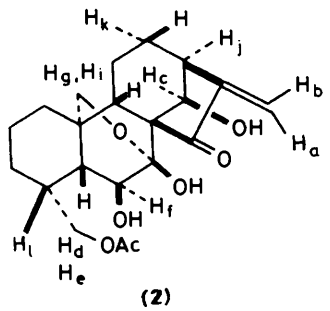


(19) R = O
(20) R = SCH_2CH_2S



(21)

was confirmed as (**1**) by chemical correlation. The less polar compound, which had the formula $C_{20}H_{28}O_5$, was presumed to have structure (**21**), which corresponds to the 1,2-didehydro-16,17-dihydro derivative of (**16**), from the fact that olefinic proton signals due to the partial structure ■–CH_2–C(H)=C(H)–■ were observed at δ 5.17 (1 H, d, *J* 10 Hz) and 5.74 (1 H, m), and other signals found for compound (**16**) were also observed in the ^{1}H n.m.r. spectrum of compound (**21**).



(2)

Longikaurin B (**2**), $[\alpha]_D$ −115.9° (C_5H_5N), was isolated as needles, m.p. 238—239.5 °C. The elemental composition was determined as $C_{22}H_{30}O_7$ from elemental analysis and the high-resolution mass spectrum. This substance also contained a five-membered-ring ketone conjugated with an exocyclic methylene group as in the case of longikaurin A (**1**), as shown from the following spectral data: u.v. [$\lambda_{max.}$(MeOH) 236 nm], i.r. [$\lambda_{max.}$(Nujol) 1 710 and 1 650 cm^{-1}], ^{1}H n.m.r. [δ(C_5D_5N) 5.51 (H_b) and 6.26 (H_a) (each 1 H, br s)], and ^{13}C n.m.r. [δ(C_5D_5N) 120.2 (t), 153.5 (s), and 209.3 (s)] (see Table 1). The ^{13}C n.m.r. spectrum of compound (**2**) (Table 1) showed, in addition, the presence of two secondary carbinyl carbons (δ_C 73.8 and 74.1), two primary carbinyl carbons (δ_C 67.2 and 67.4), and an acetal carbon (δ_C 99.0). The ^{1}H n.m.r. spectrum of (**2**) showed singlets due to a tertiary methyl group [δ 1.37 (3 H)] and an acetyl group [δ 1.95 (3 H)]. These data suggest that longikaurin B (**2**) also has the structure (**10**) as a basic skeleton. In addition, the dihydro compound (**18**) showed a negative Cotton effect in its o.r.d. spectrum. The locations of three oxygen functional groups (2 secondary hydroxy groups and 1 primary acetoxy group)

Table 3. Results of INDOR spectra and n.O.e. experiments for longikaurin B (**2**) (measured for C_5D_5N solution)

Monitor proton	Protons which showed n.O.e. for monitoring proton (%)	Protons which showed coupling with monitored proton
H_a	H_b (21)	H_b, H_j
H_b	H_a (33)	H_a, H_j
H_c	H_i	H_a, H_j
H_d	H_g (H_i), H_l	H_e
H_g H_i	H_d (11.4)	
H_j		H_c, H_k
H_l	H_d (8.0)	

were determined as follows. The two secondary hydroxy groups were located at C-14$_\beta$ and C-6$_\beta$ as in the case of longikaurin A (**1**) from the results obtained by INDOR experiments (Table 3). On monitoring the frequencies of H_a H_b, H_j [δ 3.17 (1 H, br d, *J* 10 Hz)], and H_c [5.10 (1 H, d, *J* 1 Hz)], INDOR signals were observed for H_b and H_j, H_a and H_j, H_c and H_k [δ 2.35 (1 H, m)], and H_j and H_a, respectively. Acetylation (acetic anhydride and pyridine) of longikaurin B (**2**) gave the monoacetate (**4**), m.p. 182—183 °C, in the ^{1}H n.m.r. spectrum of which H_c was observed downfield at δ 5.66 (1 H, br s) compared with its position in the spectrum of (**2**). Accordingly, H_c was assigned to 14-H_α. Another secondary carbinyl proton, H_f [δ 4.18 (1 H, d, *J* 6 Hz) on addition of D_2O] of longikaurin B (**2**), was assigned to an α-proton at C-6 on comparison of the chemical shift and the coupling pattern with those of longikaurin A (**1**). These assignments were chemically confirmed by the formation of an acetonide (**12**) and of an aldehyde lactone (**15**) on oxidation with sodium metaperiodate, as previously described. The remaining primary oxygen functional group (OAc) was presumed to be located at either C-18 or C-19 from the following facts: the number of tertiary methyl groups in longikaurin B (**2**) decreased from two to one compared with longikaurin A (**1**), and new signals [δ 4.40 and 4.68 (each 1 H, ABd, *J* 11 Hz); H_e and H_d] due to an acetoxymethyl group were observed; an n.O.e. (8%) for the proton (H_d) attached to a carbon atom bearing an acetoxy group was observed on irradiation at the frequency of the tertiary methyl group (H_l : δ 1.37). On the other hand, an n.O.e. (11.4%) was observed for H_d

on irradiation at the frequencies of H_g and H_i [δ 3.99 and 4.14 (each 1 H, ABd, *J* 10 Hz), 20-H_2]. This result shows that C-19 was oxidized to an acetoxymethyl group. Accordingly, longikaurin B has the structure *ent*-7β,20-epoxy-6α,7α,14α-trihydroxy-15-oxokaur-16-en-19-yl acetate (**2**).

Experimental

M.p.s were determined with a Yanagimoto micro melting point apparatus and are uncorrected. I.r. spectra were recorded with a Hitachi IR 215 spectrophotometer. ^{1}H N.m.r. spectra were recorded with a Hitachi R-22 (90 MHz) or JEOL PS-100 (100 MHz) spectrometer and ^{13}C n.m.r. spectra were obtained with a JEOL JNM FX 200 (50.18 MHz) spectrometer. Chemical shifts are given in δ values using tetramethylsilane as internal standard. U.v. spectra were taken with a Hitachi 124 double-beam spectrophotometer. Optical rotation and o.r.d. curves were measured with a JASCO Model ORD/UV-5 instrument. Mass spectra were determined with a JEOL JMS-01SG or a JEOL D-300 spectrometer and the values include an error within ±0.005 mass unit. Kieselgel G (0.06—0.2 mm; Merck) was used for column chromatography, and Kieselgel GF_{254} precoated plates (0.25 mm or 0.5 mm; Merck) were used for t.l.c. Extracts were dried over anhydrous magnesium sulphate.

Isolation of Longikaurin A (**1**), *B* (**2**), *and Kamebakaurin* (**5**).—The methanolic extract obtained from the dried leaves of *Rabdosia longituba* (Miq.) Hara (3.6 kg), collected on Mt. Koetsu (Tokushima Prefecture, Japan) in late August, 1977, was concentrated under reduced pressure to *ca.* 4 l, and water was added to the extract to make a 90% methanolic solution. This solution was washed with n-hexane (1.5 l × 3) and then evaporated under reduced pressure. The residue was suspended in water (3 l) and extracted with ethyl acetate (1 l × 3). After being washed with water, the extract was dried, and evaporated under reduced pressure to give a residue (169 g). The residue was chromatographed on a silica gel (2.4 kg) column with stepwise elution with chloroform–acetone as eluant (increasing acetone content). Elution with chloroform–acetone (85:15) gave first longikaurin A (**1**) and then longikaurin B (**2**). Elution with chloroform–acetone (8:2) gave kamebakaurin (**5**). The yields, physical properties, and spectral data of isolated compounds are as follows.

Longikaurin A (**1**) (3.21 g), needles, m.p. 223—225 °C (from methanol); $[\alpha]_D^{25}$ −91.1° (*c* 0.21 in pyridine); $\lambda_{max.}$(MeOH) 236 nm (ε 9 530); $\nu_{max.}$(Nujol) 3 400—3 050, 1 705, 1 640, 1 080, 1 060, and 1 020 cm^{-1}; δ($CDCl_3$) 1.09 (6 H, s, CM_2), 2.50 (1 H, m, 12-H_α), 3.03 (1 H, br d, *J* 10 Hz, 13-H), 3.60—3.88 [2 H, 3.81 (1 H, ABdd, *J* 10 and 1 Hz, 20-H) and 3.77 (1 H, d, *J* 7 Hz, 6-H) after D_2O treatment], 4.07 (1 H, ABd, *J* 10 Hz, 20-H), 4.76 (1 H, d, *J* 2 Hz, 14-H), 4.90 (1 H, m, OH), 5.52 and 6.14 (each 1 H, br s, together 17-H_2), 6.10 (1 H, m, OH), and 6.59 (1 H, d, *J* 10 Hz, OH) (Found: C, 68.8; H, 8.1. $C_{20}H_{28}O_5$ requires C, 68.94; H, 8.10%).

Longikaurin B (**2**) (1.62 g), needles, m.p. 238—239.5 °C (from chloroform–methanol); $[\alpha]_D^{25}$ −115.9° (*c* 0.12 in pyridine); $\lambda_{max.}$(MeOH) 236 nm (ε 9 150); $\nu_{max.}$(Nujol) 3 490, 3 350—3 100, 1 745, 1 710, and 1 650 cm^{-1}; δ(C_5D_5N) 1.37 (3 H, s, 18-H_3), 1.95 (3 H, s, OCOMe), 2.35 (1 H, m, 12-H_α), 3.17 (1 H, br d, *J* 10 Hz, 13-H), 3.99 and 4.14 (each 1 H, ABd, *J* 10 Hz, together 20-H_2), 4.20—4.40 [1 H, 4.18 (d, *J* 6 Hz) after D_2O treatment, 6-H], 4.40 and 4.68 (each 1 H, ABd, *J* 11 Hz, together 19-H_2), 5.10 (1 H, d, *J* 1 Hz, 14-H), 5.51 and 6.26 (each 1 H, br s, together 17-H_2), 6.92 (1 H, d, *J* 10 Hz, OH), and 7.27 (1 H, m, OH) (Found: M^+, 406.200; C, 64.9; H, 7.8%. $C_{22}H_{30}O_7$ requires *M*, 406.1992; C, 65.01; H, 7.44%).

Kamebakaurin (**5**) (2.52 g), needles, m.p. 229—230 °C (from methanol); $[\alpha]_D^{25}$ −102.4° (*c* 0.13 in pyridine); $\lambda_{max.}$(MeOH) 233 (ε 8 100); $\nu_{max.}$(Nujol) 3 550—3 200, 1 710, and 1 640 cm^{-1}; δ(C_5D_5N) 0.82 and 0.92 (each 3 H, s, together CMe_2), 3.60 (1 H, dd, *J* 10 and 5 Hz, 1-H), 4.36 and 4.66 (each 1 H, ABd, *J* 12 Hz, together 20-H_2), 4.84 (1 H, dd, *J* 10 and 6 Hz, 7-H), 5.36 (1 H, br s, 17-H), 5.62 (1 H, br s, 14-H), and 6.28 (1 H, br s, 17-H) (Found: C, 68.3; H, 8.8. Calc. for $C_{20}H_{30}O_5$: C, 68.54; H, 8.36%). This substance was found to be identical with an authentic sample of kamebakaurin (**5**)[4] by mixed m.p. and by comparison of the i.r. and ^{1}H n.m.r. spectra.

Longikaurin A 14-*Acetate* (**3**).—Acetylation of longikaurin A (**1**) (88 mg) with a mixture of acetic anhydride (1 ml) and pyridine (1 ml) gave the *monoacetate* (**3**) (47 mg) as needles, m.p. 233—235 °C (from methanol); $\nu_{max.}$(Nujol) 3 500, 3 400, 1 760, 1 710, and 1 640 cm^{-1}; δ($CDCl_3$) 1.08 and 1.10 (each 3 H, s, together CMe_2), 2.02 (3 H, s, OAc), 3.13 (1 H, br d, *J* 10 Hz, 13-H), 3.64—4.10 [3 H, 3.71 (1 H, d, *J* 6 Hz, 6-H) and 3.78 and 4.05 (each 1 H, ABd, *J* 10 Hz, together 20-H_2) after D_2O treatment], 4.12 (1 H, s, OH), 5.46 (1 H, br s, 17-H), 5.68 (1 H, d, *J* 1 Hz, 14-H), 6.03 (1 H, d, *J* 10 Hz, OH), and 6.12 (1 H, br s, 17-H); *m/z*, 390 (M^+) (Found: C, 66.4; H, 8.0. $C_{22}H_{30}O_6 \cdot \frac{1}{2}H_2O$ requires C, 66.14; H, 7.83%).

Longikaurin A 7,14-*Acetonide* (**11**).—To a solution of longikaurin A (**1**) (100 mg) in DMF (2 ml) were added 2,2-dimethoxypropane (2 ml) and PTSA (2 mg) and the mixture was refluxed gently for 6.5 h. The reaction mixture was concentrated under reduced pressure and the residue was extracted with methylene dichloride. The extract was washed successively with saturated aq. sodium chloride and water, dried, and evaporated. The residue was recrystallized from methanol to give the *acetonide* (**11**) (91 mg) as needles, m.p. 194—196 °C; $\nu_{max.}$(Nujol) 3 370, 1 715, and 1 645 cm^{-1}; δ($CDCl_3$) 1.12 and 1.15 (each 3 H, s, together 4-Me_2), 1.32 and 1.64 (each 3 H, s, together acetonide Me_2), 3.04 (1 H, br d, *J* 8 Hz, 13-H), 3.71 (1 H, ABd, *J* 10 and 2 Hz, 20-H), 4.70 (1 H, d, *J* 2 Hz, 14-H), 5.52 (1 H, br s, 17-H), 5.80 (1 H, d, *J* 12 Hz, OH), and 6.14 (1 H, br s, 17-H) (Found: M^+, 388.224; C, 71.1; H, 8.5%. $C_{23}H_{32}O_5$ requires *M*, 388.2250; C, 71.10; H, 8.30%).

Sodium Metaperiodate Oxidation of Longikaurin A (**1**).—To a solution of longikaurin A (**1**) (100 mg) in methanol (10 ml) was added a solution of sodium metaperiodate (600 mg) in water (3 ml) and the reaction mixture was stirred at room temperature for 5 days. The reaction mixture was concentrated under reduced pressure and the residue was extracted with ethyl acetate. After being washed with water and dried, the extract was evaporated under reduced pressure. The residue was separated on a silica gel (3 g) column with stepwise elution with chloroform–acetone (increasing acetone content). The eluate from chloroform gave an unstable aldehyde lactone (**13**) (25.4 mg), and the eluate from chloroform–acetone (95:5) gave a carboxylic acid lactone (**14**) (48.3 mg).

Aldehyde lactone (**13**), m.p. 145—147 °C (from diethyl ether); $\nu_{max.}$($CHCl_3$) 3 600—3 250, 1 740, 1 720, 1 710, and 1 645 cm^{-1}; δ($CDCl_3$) 1.04 and 1.16 (each 3 H, s, together CMe_2), 2.27 (1 H, d, *J* 6 Hz, 5-H), 3.11 (1 H, br d, 10 Hz, 13-H), 4.58 (1 H, br s, 14-H), 4.63 and 4.98 (each 1 H, ABd, *J* 9 Hz, together 20-H_2), 5.62 and 6.22 (each 1 H, br s, together 17-H_2), and 9.85 (1 H, d, *J* 6 Hz, 6-H) [Found: M^+ (FD), 346. M^+ (EI), 346.168. $C_{20}H_{26}O_5$ requires *M*, 346.1778].

Carboxylic acid lactone (**14**), m.p. 195—197 °C (from diethyl ether); $\nu_{max.}$(KBr) 3 650—3 300, 1 735, 1 705sh, 1 695, and 1 645 cm^{-1}; δ($CDCl_3$) 1.16 and 1.22 (each 3 H, s, together CMe_2), 2.47 (1 H, s, 5-H), 3.13 (1 H, br d, *J* 10 Hz, 13-H), 4.60 (1 H, ABd, *J* 11 Hz, 20-H), 4.62 (1 H, d, *J* 1 Hz, 14-H), 4.89 (1 H, ABd, *J* 11 Hz, 20-H), and 5.64 and 6.23 (each 1 H, br s, together 17-H_2) [Found: M^+ (FD), 362. M^+ (EI), 362.173. $C_{20}H_{26}O_6$ requires *M*, 362.1727].

Catalytic Hydrogenation of Longikaurin A (**1**).—Longikaurin A (**1**) (59 mg) was dissolved in methanol (10 ml) and platinum dioxide (5.5 mg) was added to the solution. The mixture was hydrogenated for 40 min. The catalyst was filtered off and the solvent was removed under reduced pressure to give a residue, which was separated on silica gel plates [methylene dichloride–acetone (8:2)] to give the dihydro compounds (**16**) (31.4 mg) and (**17**) (5.7 mg) as syrups.

(16R)-*Dihydro compound* (**16**), $\nu_{max.}$($CHCl_3$) 3 600—3 100 and 1 710 cm^{-1}; δ($CDCl_3$) 1.08 (6 H, s, CMe_2), 1.11 (3 H, d, *J* 9 Hz, 17-H_3), 3.08 (1 H, quintet, *J* 9 Hz, 16-H), 3.67 (1 H, dd, *J* 10 and 6 Hz, 6-H), 3.80 and 4.05 (each 1 H, ABd, *J* 10 Hz, together 20-H_2), 4.82 (1 H, br s, 14-H), 6.04 (1 H, br s, OH), and 6.37 (1 H, d, *J* 10 Hz, OH); o.r.d. $\lambda_{max.}$(MeOH) (φ): 316 (−5 141) and 280 nm (+2 981) (Found: M^+, 350.210. $C_{20}H_{30}O_5$ requires *M*, 350.2093.

(16S)-*Dihydro compound* (**17**), $\nu_{max.}$($CHCl_3$) 3 600—3 100 and 1 720 cm^{-1}; δ($CDCl_3$) 1.04 (6 H, s, CMe_2), 1.32 (3 H, d, *J* 8 Hz, 17-H_3), 3.64—3.88 (2 H, 6- and 20-H), 4.08 (1 H, ABd, *J* 10 Hz, 20-H), 4.70 (1 H, br s, 14-H), 5.66 (1 H, m, OH), and 6.38 (1 H, d, *J* 12 Hz, OH); o.r.d. $\lambda_{max.}$(MeOH) (φ): 317 (−1 312) and 279 nm (+210) (Found: M^+, 350.206).

Dithioacetalization of Compound (**19**).—To a solution of compound (**19**) (184 mg) in ethane-1,2-dithiol (0.5 ml) was added 47% boron trifluoride–diethyl ether (0.5 ml) and the reaction mixture was stirred overnight at room temperature, poured into saturated aq. sodium carbonate (50 ml), and extracted with ethyl acetate. The extract was washed with water, dried, and evaporated under reduced pressure to give a residue, which was purified on a silica gel (15 g) column with chloroform–acetone as solvent. The eluate from chloroform–acetone (95:5) gave the *dithioacetal* (**20**) (99 mg) as an amorphous powder, $\nu_{max.}$(Nujol) 3 600—3 100 and 1 705 cm^{-1}; δ(C_5D_5N) 1.09 (3 H, d, *J* 7 Hz, 17-H_3), 1.15 and 1.25 (each 3 H, s, together CMe_2), 4.16 [1 H, m (changed to d, *J* 5 Hz, after D_2O treatment), 6-H], 4.33 and 4.57 (each 1 H, ABd, *J* 10 Hz, together 20-H_2), and 5.40 (1 H, d, *J* 2 Hz, 14-H); o.r.d. $\lambda_{max.}$(MeOH) (φ): 318 (−4 882) and 288 nm (+482) (Found: M^+, 440.171. $C_{22}H_{32}O_5S_2$ requires *M*, 440.1691).

Desulphurization of Dithioacetal (**20**).—To a solution of dithioacetal (**20**) (99 mg) in ethanol (30 ml) was added Raney-nickel W-2 (2 g) and the mixture was refluxed overnight. After the mixture had cooled and been filtered, the solvent was removed under reduced pressure to give a residue (63 mg), which was separated on silica gel plates with diethyl ether as solvent. The less polar band gave 1,2-didehydrodihydrolongikaurin A (**21**) (11 mg) and the polar band gave (16*R*)-dihydrolongikaurin A (**16**).

(16*R*)-Dihydrolongikaurin A (**16**) (Found: M^+, 350.207. Calc. for $C_{20}H_{30}O_5$: *M*, 350.2093). This product was identical with an authentic sample of compound (**16**) (i.r., ^1H n.m.r.).

1,2-*Didehydro*-16,17-*dihydrolongikaurin A* (**21**), $\nu_{max.}$($CHCl_3$) 3 650, 3 600—3 050, 1 710, and 1 600 cm^{-1}; δ($CDCl_3$) 1.04 and 1.15 (each 3 H, s, together CMe_2), 1.16 (3 H, d, *J* 8 Hz, 17-H_3), 3.04 (1 H, quintet, *J* 8 Hz, 16-H), 3.72 [1 H, dd, *J* 12 and 8 Hz (changed to d, *J* 8 Hz, after D_2O treatment), 6-H], 3.82 and 3.95 (each 1 H, ABd, *J* 10 Hz, together 20-H_2), 4.90 (1 H, d, *J* 1 Hz, 14-H), 5.17 (1 H, d, *J* 10 Hz, 1-H), 5.74 (1 H, m, 2-H), and 6.02 (1 H, d, *J* 12 Hz, OH) (Found: M^+, 348.195. $C_{20}H_{28}O_5$ requires *M*, 348.1937).

Dihydrolongikaurin B (**18**).—Longikaurin B (**2**) (70 mg) was dissolved in methanol (20 ml) and platinum dioxide (3 mg) was added to the solution. The mixture was hydrogenated (1 h) and the catalyst was filtered off. After removal of the solvent the residue was purified on silica gel plates [methylene dichloride–acetone (8:2)] to give *dihydrolongikaurin B* (**18**) (40 mg) as a syrup, $\nu_{max.}$($CHCl_3$) 3 600—3 100, 1 730, and 1 720 cm^{-1}; δ(C_5D_5N) 1.13 (3 H, d, *J* 6 Hz, 17-H_3), 1.36 (3 H, s, 18-H_3), 1.96 (3 H, s, OAc), 3.28 (1 H, quintet, *J* 8 Hz, 16-H), 4.00 (2 H, br s, 20-H_2), 4.28 [1 H, m (changed to d, *J* 6 Hz after D_2O treatment), 6-H], 4.38 and 4.65 (each 1 H, ABd, *J* 10 Hz, together 19-H_2), and 5.10 (1 H, d, *J* 2 Hz, 14-H); o.r.d. $\lambda_{max.}$(MeOH) (φ): 317 (−4 004) and 280 nm (+3 022) (Found: M^+, 408.210. $C_{22}H_{32}O_7$ requires *M*, 408.2148).

Longikaurin B 14-*Acetate* (**4**).—Longikaurin B (**2**) (54 mg) was acetylated with a mixture of acetic anhydride (1 ml) and pyridine (1 ml). The reaction product was recrystallized from dil. aq. ethanol to give *longikaurin B* 14-*acetate* (**4**) (26 mg) as needles, m.p. 182—183 °C; $\nu_{max.}$(Nujol) 3 500—3 100, 1 740, 1 705, 1 650, and 1 240 cm^{-1}; δ($CDCl_3$) 1.20 (3 H, s, Me), 2.40 and 2.06 (each 3 H, s, OAc), 3.17 (1 H, br d, *J* 10 Hz, 13-H), 3.76—4.20 (4 H, m, 6-H, 19-H, and 20-H_2), 4.16 (1 H, s, OH), 4.34 (1 H, ABd, *J* 12 Hz, 19-H), 5.48 (1 H, br s, 17-H), 5.66 (1 H, br s, 14-H), 6.10 (1 H, d, *J* 12 Hz, OH), and 6.14 (1 H, br s, 17-H) (Found: C, 64.1; H, 7.3. $C_{24}H_{32}O_8$ requires C, 64.27; H, 7.19%).

Longikaurin B 7,14-*Acetonide* (**12**).—Longikaurin B (**2**) (53 mg) was dissolved in DMF (1 ml), and 2,2-dimethoxypropane (1 ml) and PTSA (1 mg) were added to the solution. The reaction mixture was refluxed gently overnight and treated as previously described for compound (**11**). The product was purified on silica gel plates [methylene dichloride–acetone (9:1)] to give the *acetonide* (**12**) (16.6 mg) as a syrup $\nu_{max.}$($CHCl_3$) 3 500—3 250, 1 730, 1 720, and 1 645 cm^{-1}; δ($CDCl_3$) 1.27 (3 H, s, 4-Me), 1.34 and 1.64 (each 3 H, s, together acetonide Me_2), 2.08 (3 H, s, OAc), 3.07 (1 H, br d, *J* 10 Hz, 13-H), 3.64—4.24 (4 H, m, 19-H, 20-H_2, and 6-H), 4.51 (1 H, ABd, *J* 10 Hz, 19-H), 4.70 (1 H, d, *J* 2 Hz,14-H), 5.54 and 6.16 (each 1 H, br s, together 17-H_2), and 5.82 (1 H, d, *J* 12 Hz, OH) (Found: M^+, 446.228. $C_{25}H_{34}O_7$ requires *M*, 446.2305).

Sodium Metaperiodate Oxidation of Longikaurin B (**2**).—To a solution of longikaurin B (**2**) (50 mg) in methanol (10 ml) was added a solution of sodium metaperiodate (500 mg) in water (4 ml), and the reaction mixture was stirred at room temperature for 5 days, then was treated as previously described for the oxidation of compound (**1**), and the product was purified on a silica gel (3 g) column with stepwise elution with chloroform–acetone. The eluate from chloroform–acetone (95:5) gave the unstable *aldehyde lactone* (**15**) (13.1 mg) as a syrup, $\nu_{max.}$($CHCl_3$) 3 600—3 275, 1 740, 1 705, and 1 645 cm^{-1}; δ($CDCl_3$) 1.17 (3 H, s, Me), 2.05 (3 H, s, OAc), 2.31 (1 H, d, *J* 4 Hz, 5-H), 3.12 (1 H, br d, *J* 8 Hz, 13-H), 4.03 and 4.30 (each 1 H, ABd, *J* 12 Hz, together 19-H_2), 4.60 (1 H, d, *J* 1 Hz, 14-H), 4.76 (2 H, br s, 20-H_2), 5.64 and 6.25 (each 1 H, br s, together 17-H_2), and 9.86 (1 H, d, *J* 4 Hz, 6-H) [Found: M^+ (FD), 404. M^+ (EI), 404.182. $C_{22}H_{28}O_7$ requires *M*, 404.1832].

Acknowledgements

We wish to express our sincere thanks to Mr. G. Murata, Faculty of Sciences, Kyoto University, for identification of plant material and to the staff of the Analytical Centre of this Faculty for the n.m.r. spectra, mass spectra, and elemental analyses.

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Received 16*th February*, 1987; *Paper* 7/282