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CHEMICAL STUDIES OF ORIENTAL PLANT DRUGS—XXXIV

DITERPENES OF *ARALIA CORDATA*: OXIDATIVE TRANSFORMATION OF 4-AXIAL ALDEHYDE OF SOME DITERPENES AND A NOTE TO THE NATURALLY OCCURRING 4-HYDROXY-18(or 19)NORDITERPENES

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Abstract— It has been found that the 4-axial aldehyde of the diterpenes (VIII. XII and XVI) is labile; a concentrated solution being readily autoxidized at room temperature. The structures of the autoxidation products. the reaction mechanism and the solvent effect have been elucidated.

Accordingly. 4-hydroxy-18(or 19)norditerpenes were prepared. Some 4-hydroxynorditerpenes which have been reported as being isolated from natural sources are probably artifacts derived from the corresponding 4-axial aldehyde compounds.

THE isolation and the structural elucidation of the (−)-pimaradiene type diterpenes. I, II, III and IV of *Aralia cordata* Thunb. (Japanese name: Udo), one of the original plants of Chinese drug "Duhuo" have been reported.[2]

(−)-Kaurenoic acid (V) and its related diterpene (VI) have also been isolated from the same plant.

In addition, other new diterpenes, (−)-primara-8(14),15-dien-19-ol (VII) and (−)-pimara-8 (14),15-dien-19-al (VIII) have been obtained from the neutral fraction of the ethereal extracts of the roots, the latter of which was found to be quite unstable and its structure was finally established by the comparison with the aldehyde prepared from VII as the semicarbazone m.p. 228–229°.§ It has been reported that an axial aldehyde at C-4 of some of the diterpenes is very unstable and is readily

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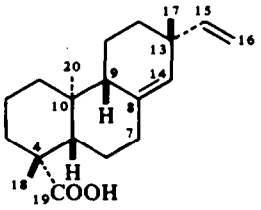
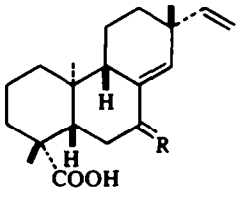
§ Hanson and White have prepared this semicarbazone during the course of the study of gibberellin biosynthesis (J. Chem. Soc. (C), 981 (1969)). They reported m.p. 187–188°, but no analytical and spectral data were described in their paper.

CHART 1



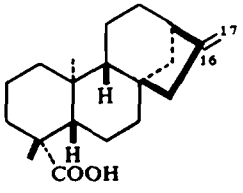
I



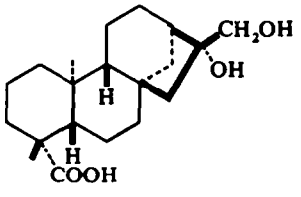
II: R = 

III: R = 

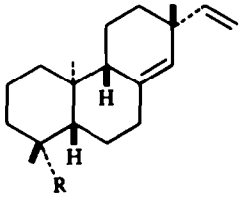
IV: R = =O



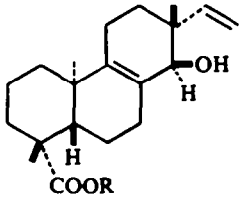
V



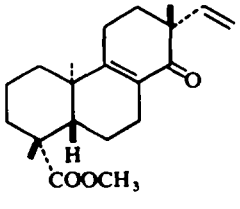
VI



VII: R = CH_2OH
VIII: R = CHO



IX: R = H
X: R = CH_3

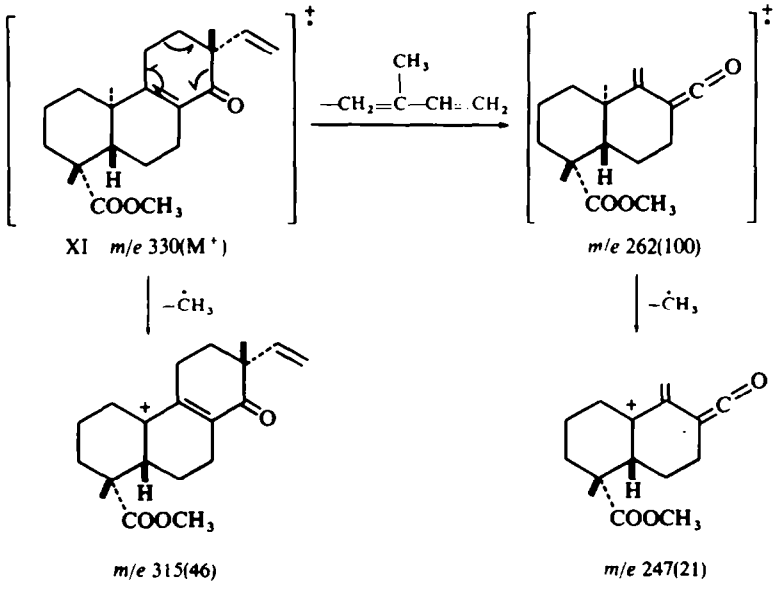


XI

decomposed in solution.[3] However, the separation and the characterization of the products have not so far been investigated. The present paper describes oxidative decomposition of such aldehydes and is significant for the synthesis and biogenesis of 4-hydroxy-18(or 19)-norditerpenes which recently have been found in nature.

A solution of VIII in benzene was allowed to stand overnight at room temperature. The column chromatography of the crude products on silica gel afforded two crystalline compounds, one of which was identical with the parent acid (I). Another compound (IX), a hydroxy acid, $C_{20}H_{30}O_3$, IRν_{max}^{KBr} 3386 and 1690 cm^{-1}, gave a methyl ester (X), $\nu_{max}^{CCl_4}$ 3628 and 1728 cm^{-1}. The NMR and IR spectra of IX and X indicated that the vinyl group remained unreacted but the trisubstituted double bond was absent in IX and X. The NMR signal of IX (in pyridine-d_5) at δ 4·03 (1H, singlet) and that of X (in $CDCl_3$) at δ 3·79 (1H, singlet) revealed the presence of a secondary OH group having no proton on its α-C atoms. Mild oxidation of X afforded an α,β-unsaturated ketone (XI), IR$\nu_{max}^{CCl_4}$ 1733 and 1677 cm^{-1}, UVλ_{max}^{EtOH} 249·5 mμ (ε = 7200), showing the allylic nature of this OH group. These observations led to the formulation of IX as 14-hydroxy-(−)-pimara-8,15-dien-19-oic acid which was further supported by the mass spectrum of XI as illustrated in Chart 2.[4] Although the configuration of the OH group has not been defined, it should be β on the basis of the mechanism of the formation of IX from VIII as mentioned later.

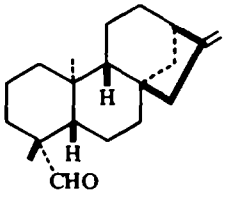
CHART 2.



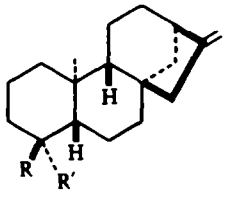
Subsequently the autoxidation of (−)-kaur-16-en-19-al (XII) prepared from V was examined. On standing at room temperature for 2 days, a solution of XII in benzene yielded a complex mixture, from which V and other two compounds, XIII and XIV, were isolated. The positive NaI test and the IR absorption at 3559 cm^{-1} (in CCl_4)[5] as well as the mass and NMR spectra indicated that XIII and XIV can be represented by a pair of epimers of 4-hydroperoxy-18(or 19)-nor-(−)-kaur-16-ene. The respective formulation of XIII and XIV as 4β and 4α-hydroperoxide was reasonably in accord with the assignments of NMR Me signals of XIII at δ 1·11 to 19-Me and at δ 1·03 to 20-Me and those of XIV at δ 1·13 to 18-Me and at δ 1·29 to 20-Me, because the 4α(axial)-hydroperoxyl group of XIV was expected to result in the downfield shift

of 20-Me resonance (*vide infra*). Although the complete purification of other products could not be achieved owing to their complexity and their further decomposition during the column chromatography on silica gel, the NMR spectra of the crude materials suggested that the additional products would be formed not only by the autoxidation of the aldehyde but also by the concomitant degradation of the double bond. In order to avoid such a complication, the autoxidation of the aldehyde compound having no double bond was investigated.

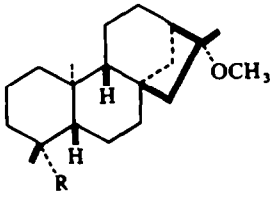
CHART 3



XII



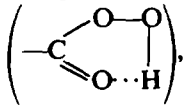
XIII: R = OOH, R' = CH_3
XIV: R = CH_3, R' = OOH



XV: R = COOH
XVI: R = CHO
XVII: R = $COOCH_3$
XVIII: R = CH_2OH

Treatment of V with H_2SO_4 in MeOH afforded 16 α-methoxy-(−)-kauranoic acid (XV),[6] which was converted to 16 α-methoxy-(−)-kauran-19-al (XVI) via the methyl ester (XVII) and the alcohol (XVIII). A benzene solution of XVI at various concentrations—0·2, 1·0, 5·0 and 25·0 %—was allowed to stand at room temperature and the decomposition process of each solution was followed by TLC. As shown in Fig 1, the constitution of the products was found to depend upon the concentration of the solutions. In case of the 25% solution, the decomposition occurred very fast in the early stage but some of the aldehyde (XVI) still remained unchanged even after 3 days. This would be due to the deficiency of oxygen in benzene which was not enough to oxidize all the substrate at such a high concentration.

From the mixture of the 1% solution, compounds XIX and XX were isolated, the former was idenfified as the parent acid (XV). The latter (XX), $C_{21}H_{34}O_4$, which was a major product in case of decomposition of 0·1 and 1·0% solutions, was proved to be the peracid of XV on the basis of IR spectrum, $\nu_{max}^{CCl_4}$ 3253 and 1732 cm^{-1}



, as well as NMR spectrum and the elemental analysis.

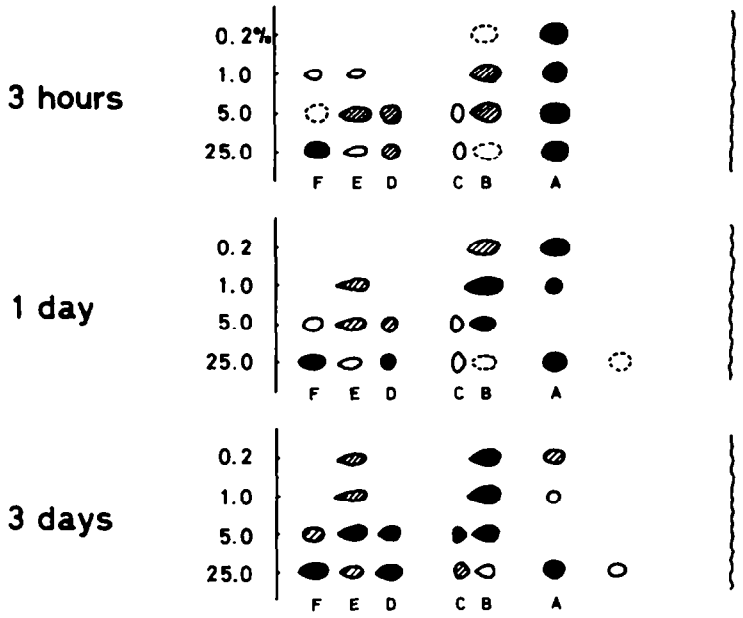


FIG 1. Thin layer chromatograms of autoxidation products of XVI
Solvent: benzene–EtOAc–10:1
A: XVI. B: XX. C: XXI. D: XXII. E: XIX=XV. F: XXIII

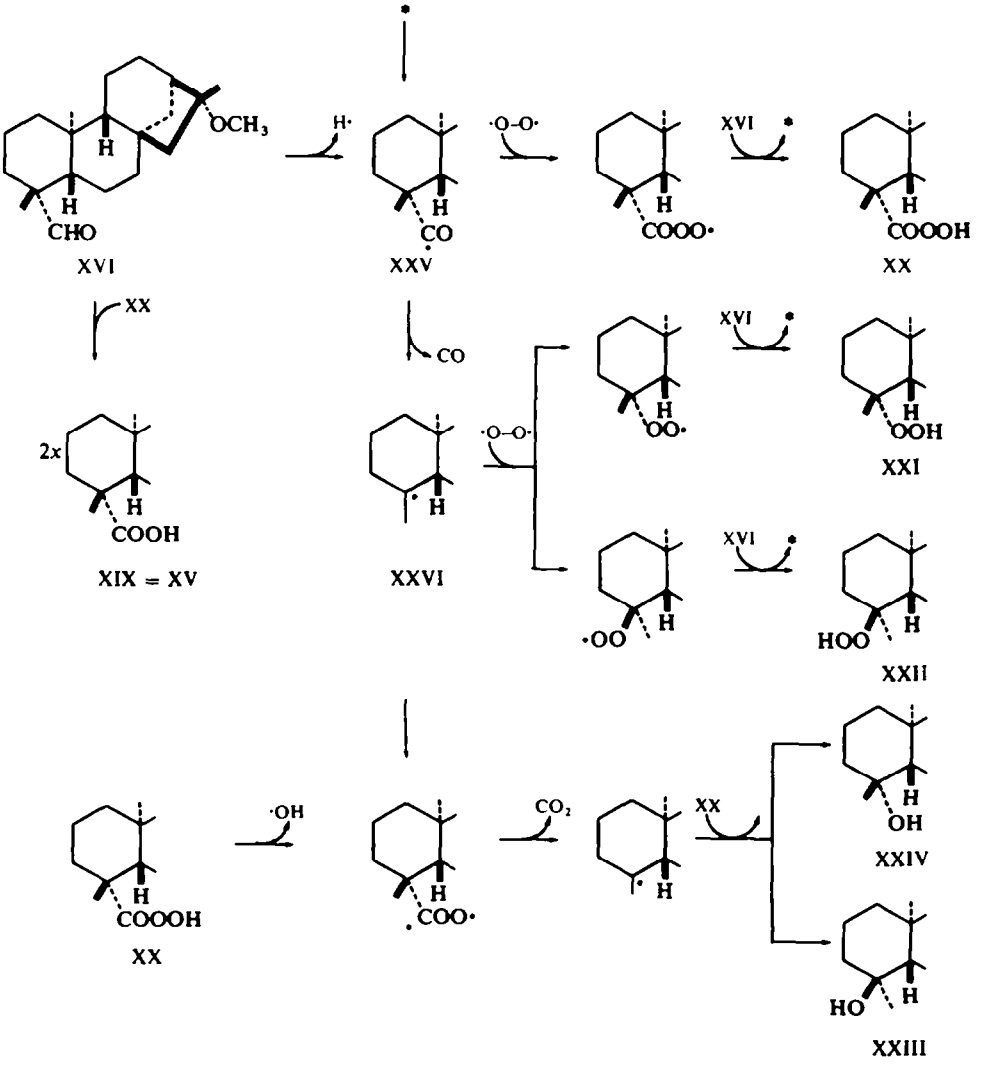
The compounds XXI, $C_{19}H_{31}O_2(OCH_3)$, XXII, $C_{19}H_{31}O_2(OCH_3)$, and XXIII, $C_{19}H_{31}O(OCH_3)$, were obtained from the mixture of a conc solution together with the acid XIX. The IR, NMR and mass spectra indicated that XXI and XXII are a pair of epimers of 4-hydroperoxy-16α-methoxy-18(or19)-nor-(−)-kaurane. On reduction with NaI, XXI yielded an alcohol (XXIV), $C_{19}H_{31}O(OCH_3)$, while XXII afforded an isomeric alcohol, which was identical with XXIII. It has been well known that an OH group gives rise to the remarkable downfield shift of the NMR signal of the Me group at the 1,3-diaxial or geminal relation, and this effect is significantly emphasized in pyridine.[8] As shown in Table 1, the large solvent effect of pyridine was observed in the two Me signals of XXIV, whereas in only one of XXIII showing that XXIV should be the 4α(axial)-hydroxy-19-nor compound and consequently XXIII can be formulated as 4β(equatorial)-hydroxy-18-nor diterpene. Therefore, the hydroperoxyl group of XXI and XXII can be assigned as 4α and 4β, respectively. It should be noted that the 4α(axial)-hydroperoxyl group like the 4α(axial)-hydroxyl group causes the marked downfield shift of the 20-Me signal. This effect coincides with the structural assignment of XXI and XXII as mentioned above.

The peracid (XX) is stable in benzene solution at a concentration below 1·0%, but extremely unstable in the crystalline state or in a conc solution decomposing rapidly to yield XXIII and a trace of XXIV,[9] whereas, the hydroperoxides XXI and XXII were stable even in a conc solution. This evidence, coupled with the generally accepted autoxidation sequence of an aldehyde support the mechanism of the formation of

TABLE 1. NMR METHYL SIGNALS OF AUTOXIDATION PRODUCTS OF XVI

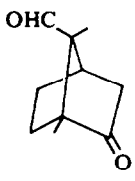
Compound	Solvent	20-Me	18(19)-Me	17-Me	O-Me
XIX = XV	$CDCl_3$	0·94	1·21	1·26	3·11
XX	$CDCl_3$	0·86	1·24	1·26	3·11
XXI	$CDCl_3$	1·26	1·11	1·26	3·11
XXII	$CDCl_3$	1·02	1·10	1·28	3·13
XXIII	$CDCl_3$	0·98	1·11	1·27	3·12
	pyridine-d_5	0·99	1·24	1·28	3·09
XXIV	$CDCl_3$	1·17	1·17	1·29	3·13
	pyridine-d_5	1·42	1·33	1·28	3·12

CHART 4

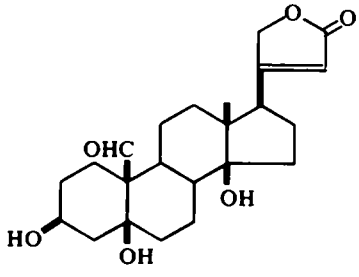


XIX, XX, XXI, XXII and XXIII from XVI as illustrated in Chart 4. A formyl radical (XXV) initially formed from XVI is immediately attacked by the oxygen dissolved in the solution to give the peracid (XX). In the concentrated solution, however, the oxygen would be rapidly consumed, and prior to the oxygen attack, the excess radical (XXV) is decomposed to a radical (XXVI) with elimination of CO, which affords the hydroperoxides XXI and XXII. This inference is supported by the fact that the 5% solution gave the only XIX and XX under aeration. The replacement of a tertiary aldehyde group by a tertiary OH group under such a mild condition has already been reported in the auto-decomposition of *trans*-π-oxocamphor (XXVII),[10] strophanthidin

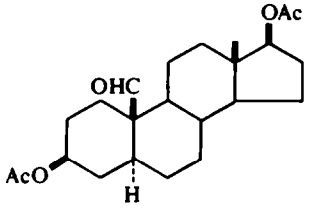
CHART 5



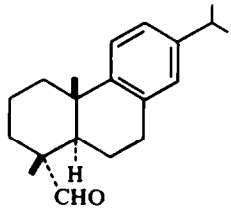
XXVII



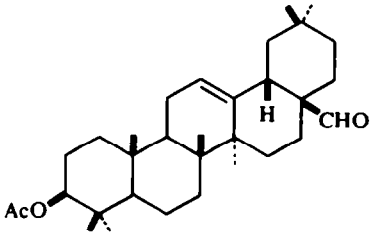
XXVIII



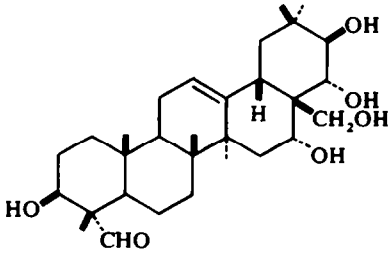
XXIX



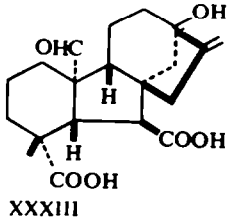
XXX



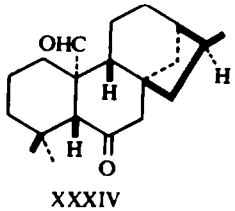
XXXI



XXXII



XXXIII

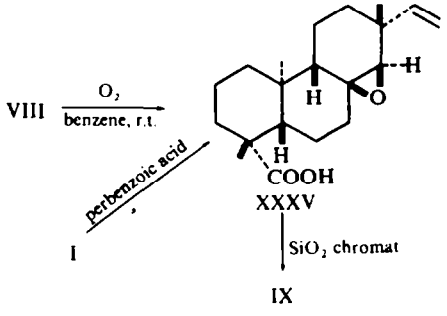


XXXIV

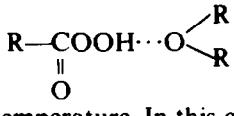
(XXVIII)[11] and some other 10-formyl-steroids (XXIX),[12] but the present study is the first example of the isolation and the identification of the peracid and the hydroperoxides, the former being an intermediate of the OH compound. It should be noted that this facile autoxidation does not always occur. Dehydroabietinal(=(+)-abieta-8,11,13-trien-18-al) (XXX), 3β-acetoxy-olean-12-en-28-al (XXXI),[13] theasapogenol E (XXXII),[14] gibberellin A_{19} (XXXIII)[15] and the aldehyde group at C-10 of kaurene type diterpenes (XXXIV)[16] are quite stable under such conditions.

The oxidative decomposition of XVI has suggested that the primary product of the autoxidation of the aldehyde (VIII) is not IX but probably the epoxide (XXXV). It has been reported that the oxidation of pimaric acid(=(+)-pimara-8(14),15-dien-18-oic acid) with the peracid yielded selectively the 8(14)-monoepoxide and the epoxide of this type was highly reactive.[17] On careful oxidation with perbenzoic acid, the acid (I) afforded a crystalline compound which was proved to be the 8β(14β)-monoepoxide (XXXV)[18] by the NMR spectrum though the complete purification could not be achieved owing to its instability as well as to the contamination by a trace of the diepoxide. The column chromatography of this epoxide (XXXV) on silica gel caused the opening of the epoxide ring eluting a compound which was identical with IX. Although the R_f value and colour reaction (with H_2SO_4) of XXXV on the thin layer chromatogram were similar to those of IX, the careful comparison of the crude autoxidation mixture of VIII with XXXV and IX by the thin layer chromatography proved the presence of XXXV and the absence of IX in the products prior to the column chromatography, indicating that IX must be the secondary product formed from XXXV during development of the column chromatography. The process of the autoxidation of VIII in benzene can be now summarized as shown in Chart 6.

CHART 6



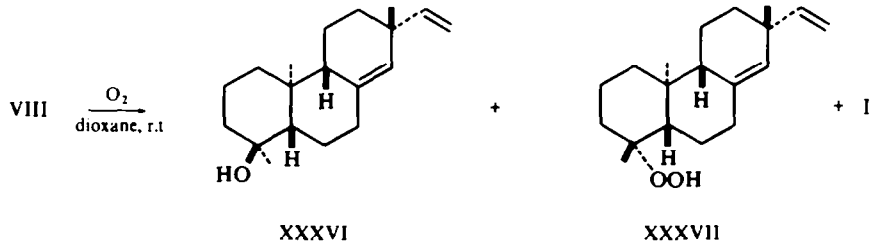
It has recently been found that epoxidation of a double bond with an organic peracid in a solvent having basic oxygen such as dioxan proceeds very slowly because of the H-bond formation between a peracid and the solvent [19]



A solution of VIII in dioxan was allowed to stand at room temperature. In this case, the decomposition proceeded more slowly than in benzene and from the mixture, two crystalline compounds XXXVI (30%) and XXXVII (3%) along with the acid (I) were isolated. The formation of the epoxide (XXXV) was excluded by TLC. On comparison

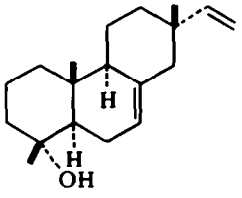
of the NMR, IR and mass spectra with those of XXIII and XXIV, XXXVI was assigned as 4β-hydroxy-19-nor(−)-pimara-8(14),15-diene.* Although XXXVII has not been completely purified, the IR and NMR spectra as well as the positive NaI test were consistent with the formulation of this compound as 4α-hydroperoxy-18-nor-(−)-pimara-8(14),15-diene. This result revealed that the autoxidation of the 4-axial aldehyde in solution was remarkably affected by the solvent and under some conditions the decomposition occurred without any oxidation of the double bond.

CHART 7

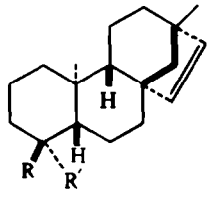


XXXVI XXXVII

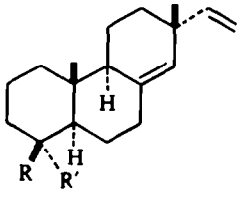
CHART 8



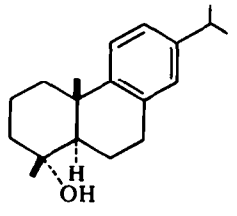
XXXVIII



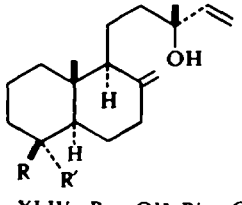
XXXIX: R = CH_3, R' = OH
XL: R = OH, R' = CH_3 (and its epoxide)



XLI: R = OH, R' = CH_3
XLII: R = CH_3, R' = OH



XLIII



XLIV: R = OH, R' = CH_3
XLV: R = CH_3, R' = OH
XLVI: R = CHO, R' = CH_3

* Very recently, the antipode of XXXVI, m.p. 119–121°, $[\alpha]_D$ +92°, has been isolated from *Pinus contorta* (J. W. Rowe, R. C. Ronald and B. A. Nagasampagi, Phytochem., **11**, 365 (1972).

Recently the occurrence of 4-hydroxy-19(or 18)-norditerpenes such as XXXVIII,[20] XXXIX, XL,[21] XLI, XLII,[22] XLIII[23] have been reported. However, the present study strongly suggests that some would be the secondary products formed from the corresponding 4-axial aldehyde during the course of isolation. Very recently Capto *et al.*[24] reported that the norditerpenes XLIV and XLV of *Araucaria excelsa* are artifacts formed from the aldehyde (XLVI), though in their short communication, the details of the autoxidation mechanism of the aldehyde, has not been described.

Our present study also indicates a more efficient method of preparation of the 4-hydroxy norditerpenes, which have hitherto been derived from the corresponding acid or alcohol only in very poor yields.[3b]

EXPERIMENTAL

All m.p. were determined on a micro hot-stage and were uncorrected. The NMR spectra were measured with a JEOL's JNM 4H-100 (100 MHz) using TMS as internal standard. The chemical shifts are shown in δ value (ppm). Chromatoplates for TLC were made by the usual method using silica gel D-5 (Camag).

Isolation of (−)-pimara-8(14). 15-dien-19-ol (VII) *and (−)-pimara-8(14), 15-dien-19-al* (VIII) *from the roots of Aralia cordata.* The ethereal extract of the roots was treated with $NaHCO_3$ aq, $NaCO_3$ aq and NaOH aq and finally with water to remove the acidic constituents.[4] The remaining non-acidic fraction was chromatographed on silica gel using hexane–benzene (1:1) as a developing solvent. The first eluted substance was very unstable aldehyde and treated with semicarbazide hydrochloride to form a semicarbazone as colourless plates (from MeOH), m.p. 228–229° (dec.),*** $[\alpha]_D^{25}$ −77·3° (c = 0·35, pyridine). (Found: C, 73·12; H, 9·64; N, 12·30. Calcd. for $C_{21}H_{33}ON_3$: C, 73·42; H, 9·68; N, 12·23%). IR ν_{max}^{KBr} 3478, 1686, 1582, 997, 912, 864, 847 cm^{-1}. NMR (pyridine-d_5): 0·67, 1·04, 1·07 (singlets, 3H each, $-\overset{|}{\underset{|}{C}}-CH_3$), 5·27 (1H, broad singlet, $-\overset{|}{C}=CH-$), 5·02–5·96 (3H ABC-type multiplet, $-CH=CH_2$), 7·14 (2H broad singlet, $-NH_2$), 7·63 (1H singlet overlapped with the signal of pyridine in the solvent, $-CH=N-$), 11·02 (1H broad singlet, $-NH-$). This semicarbazone was identified as the semicarbazone of VIII by mixed fusion and IR spectrum. Further elutions with benzene and CH_2Cl_2 gave two crystalline compounds, m.p. 109–110° and m.p. 139–140°. The former, colourless needles (from acetone), $[\alpha]_D^{25}$ − 78·9° (c = 0·60, $CHCl_3$), IR ν_{max}^{KBr} 3360, 1635, 912, 861 and 847 cm^{-1}, NMR ($CDCl_3$): 0·72 (3H singlet, $-\overset{|}{\underset{|}{C}}-CH_3$), 0·98 (6H singlet, 2 · $-\overset{|}{\underset{|}{C}}-CH_3$), 3·42 and 3·87 (1H each, a pair of doublets, $-CH_2-O-$), 5·12 (1H broad singlet, $-\overset{|}{C}=CH-$) and 4·80–5·85 (3H ABC-type multiplet, $-CH=CH_2$), was identical with VII[2] by mixed fusion, TLC, IR and NMR spectra. The latter giving a positive Liebermann-Burchard reaction was supposed to be a mixture of phytosterols by TLC, IR and NMR spectra.

Preparation of (−)-pimara-8(14),15-dien-19-al (VIII). To a suspension of CrO_3 (2·00 g) in pyridine (25 ml) was added VII (1·92 g) in pyridine (25 ml) and the mixture was stirred at room temp for 4 hrs. After decomposing the excess of reagent with MeOH (40 ml), the mixture was poured into 1% KOH soln and extracted with ether. The crude product was chromatographed on silica gel using hexane–benzene (3:1) as a developing solvent to give the VIII (600 mg). The aldehyde (VIII) being very unstable was rapidly recrystallized from MeOH as colourless needles, m.p. 77–79°, $[\alpha]_D^{24}$ − 79·5° (c = 0·66, $CHCl_3$). IR ν_{max}^{KBr} 1714, 1656, 876 cm^{-1}. NMR ($CDCl_3$): 0·61, 1·01, 1·04 (singlets, 3H each, $-\overset{|}{\underset{|}{C}}-CH_3$), 5·25 (1H

broad singlet, —C=CH—), 4·86–5·93 (3H ABC-type multiplet, —CH=CH_2), 9·87 (1H singlet, —CHO).

Autoxidation of VIII *in benzene.* A solution of VIII (841 mg) in benzene (17 ml) was allowed to stand overnight at room temp. After evaporation of the solvent, the residue (957 mg) was chromatographed on silica gel using benzene–CH_2Cl_2 as a developing solvent to elute I (110 mg), which was identical with authentic sample by mixed fusion, TLC and IR spectrum.

The next eluate was recrystallized from acetone to give a hydroxy acid (IX) as colourless needles (121 mg), m.p. 219–220°, $[\alpha]_D^{25}$ − 120·4° (*c* = 0·66, pyridine). (Found: C, 75·26; H, 9·44. Calcd. for $C_{20}H_{30}O_3$: C, 75·43; H, 9·50%). NMR (pyridine-d_5): 1·23, 1·33, 1·36 (singlets, 3H each, —C(—CH_3)—), 4·03 (1H broad singlet, —CH—O—), 5·02–6·33 (3H ABX-type multiplet, —CH=CH_2).

TLC (solvent: benzene–EtOAc = 19:1 or benzene–$CHCl_3$ = 5:1) revealed that the initial products of this autoxidation must be XXXV (*vide infra*) and I, and no IX was detected.

This autoxidation was inhibited by the presence of t-butyl-4-hydroxyanisole.

Methylation of IX. The acid IX was methylated with ethereal diazomethane in the usual manner to give X as colourless needles (from acetone), m.p. 104–105°, $[\alpha]_D^{16.5}$ − 36·9° (*c* = 0·79, $CHCl_3$). (Found: C, 75·95; H, 9·85. Calcd. for $C_{21}H_{32}O_3$: C, 75·86; H, 9·70%). NMR ($CDCl_3$): 0·80, 0·95, 1·18 (singlets, 3H each, —C(—CH_3)—), 3·63 (3H singlet, —$COOCH_3$), 3·97 (1H broad singlet, —CH—O—), 4·85–5·97 (3H ABX-type multiplet, —CH=CH_2).

Oxidation of X *with chromium trioxide in pyridine.* To a suspension of CrO_3 (230 mg) in pyridine (2 ml) was added a soln of X (226 mg) in pyridine (4 ml) and the mixture was stirred at room temp for 12 hr. MeOH (4·5 ml) was then added and stirring was continued for additional 2 hr. Dilution of the mixture with water (60 ml) followed by extraction with ether (5 × 100 ml) yielded a crude product (196 mg), which was chromatographed on silica gel using hexane–benzene as a developing solvent to give XI (46 mg) as colourless plates (from MeOH), m.p. 108–109°, $[\alpha]_D^{25}$ − 53·0° (*c* = 0·92, $CHCl_3$). (Found: C, 76·43; H, 9·11. Calcd. for $C_{21}H_{30}O_3$: C, 76·32; H, 9·15%). NMR ($CDCl_3$): 0·91, 1·17, 1·24 (singlets, 3H each, —C(—CH_3)—), 3·69 (3H singlet, —$COOCH_3$), 4·92–6·29 (3H ABX-type multiplet, —CH=CH_2).

Epoxidation of I. To a soln of I (130 mg) in $CHCl_3$ (1 ml) was added 6% perbenzoic acid soln in $CHCl_3$ (0·6 ml) and the soln was kept at 4°. A perbenzoic acid soln of the same concentration was further added after 24 (2·3 ml) and 96 hrs (3·5 ml). The mixture was allowed to stand at 4° for total 6 days and then concentrated to dryness at low temperature *in vacuo*. Recrystallization of the residue from hexane or acetone gave XXXV as colourless prisms, m.p. 163–164° (38 mg), IR ν_{max}^{KBr} 1695 cm^{-1} (COOH). This epoxide (XXXV) was homogeneous by TLC (solvent: benzene–EtOAc = 19:1) but the result of the elemental analysis as well as the presence of a weak peak at *m/e* 334 (M^+ + 16) in its mass spectrum suggested the contamination of a small amount of impurity such as the diepoxide. The NMR spectrum ($CDCl_3$) which showed the main signals at 0·81, 1·07, 1·24 (singlets, 3H each, —C(—CH_3)—), 2·78 (1H singlet, —C(O)—CH—), and 4·9–6·0 (3H ABC-type multiplet, —CH=CH_2), also exhibited the weak signals due to the impurity.

The epoxide (XXXV) was quite unstable and treatment of the crude mixture with $NaHCO_3$ aq and then dilute mineral acid resulted in partial decomposition. Further, the column chromatography of XXXV on silica gel did not elute XXXV, affording colourless needles (from hexane), which was proved to be IX by mixed fusion, TLC, IR and NMR spectra.

Autoxidation of VIII *in dioxane.* A soln of the freshly prepared VIII (300 mg) in dioxane (6 ml) was allowed to stand at 20° for 6 days. The soln was concentrated to dryness *in vacuo*. TLC (solvent: benzene–EtOAc = 19:1) of the crude products indicated the formation of I, XXXVI and a small amount of XXXVII. The column chromatography of the mixture on silica gel followed by washing with alkali to remove I gave XXXVI (80 mg) as colourless needles (from aqueous acetone), m.p. 120–121·5°), $[\alpha]_D^{22}$ – 85·5 (*c* = 1·01, $CHCl_3$). (Found: C, 78·71; H, 11·35. Calcd. for $C_{19}H_{30}O.H_2O$: C, 78·15; H, 11·05%). Mass spectrum: M^+ *m/e* 274. IR $\nu_{max}^{CCl_4}$ 3610 cm^{-1} (OH) and no carbonyl and carboxyl band. NMR ($CDCl_3$): 0·69, 0·99, 1·16 (singlets, 3H each, $-\overset{|}{\underset{|}{C}}-CH_3$), 5·15 (1H broad singlet, $-C=CH-$), 4·80–5·85 (3H ABC-type multiplet, $-CH=CH_2$).

Compound XXXVII could not be recrystallized owing to the shortage of material. The crude XXXVII, m.p. 90–92°, showed the IR band (CCl_4) at 3550 cm (OOH) and NMR signals at δ 0·83, 0·98, 1·28 (singlets, 3H each, $-\overset{|}{\underset{|}{C}}-CH_3$), 5·12 (1H broad singlet, $-C=CH-$), 4·78–5·85 (3H ABC-type multiplet, $-CH=CH_2$). This compound reduces NaI to produce I_2[25].

The autoxidation of VIII in dioxane was found to be remarkably accelerated by bubbling air.

Preparation of (−)-kaur-16-en-19-al (XII) *from* V. Methyl ester of V was reduced with LAH4 to the corresponding alcohol as colourless needles, m.p. 141–142° (lit.[26], m.p. 141–142°). A soln of this alcohol (1·82 g) in pyridine (8 ml) was added to a suspension of CrO_3 (2·0 g) in pyridine (16 ml) and the mixture was stirred at room temp for 4 hr. After working up in the usual way, the product was chromatographed on silica gel using hexane–benzene as a developing solvent to give a pure sample of XII (785 mg), IR ν_{max}^{KBr} 1714, 1656 and 876 cm^{-1}, which was immediately employed for the autoxidation.

Autoxidation of XII. A soln of XII (2·30 g) in benzene (32 ml) was allowed to stand at room temp for 2 days. After evaporation of the solvent, the residue showing several spots on TLC (solvent: benzene or benzene–acetone = 4:1) was chromatographed on silica gel. The benzene fraction afforded two crystalline hydroperoxides (XIII and XIV) which showed a positive NaI test.[26]

The hydroperoxide (XIII) was recrystallized from hexane as colourless prismatic needles, m.p. 135–137°. (Found: C, 78·04; H, 10·62. Calcd. for $C_{19}H_{30}O_2$: C, 78·57; H, 10·41%). (molecular weight: Calcd. 290·4. Found by mass spectrometry, M^+ *m/e* 290). NMR ($CDCl_3$): 1·13, 1·29 (singlets, 3H each $-\overset{|}{\underset{|}{C}}-CH_3$), 4·78 (2H broad singlet, $-\overset{|}{C}=CH_2$).

The other hydroperoxide (XIV) was obtained in a very poor yield, mass spectrum: M^+ *m/e* 290 corresponding to the molecular formula $C_{19}H_{30}O_2$. NMR ($CDCl_3$): 1·03, 1·11 (singlets, 3H each, $-\overset{|}{\underset{|}{C}}-CH_3$), 4·80 (2H broad singlet, $-\overset{|}{C}=CH_2$).

The benzene–CH_2Cl_2 (1:1) fraction was chromatographed again on silica gel using benzene as a developing solvent to give a crystalline compound (100 mg), which was identical with V by mixed fusion, TLC and IR spectrum.

The more polar fractions eluted with CH_2Cl_2 and CH_2Cl_2–acetone showed to contain several substances, which could not be separated as yet.

16α-Methoxy-(−)-kauran-19-oic acid (XV). To a 3% soln (22 ml) of H_2SO_4 in MeOH was added V (219 mg) and the mixture was refluxed for 1 hr. After dilution with water, the product deposited was repeatedly extracted with ether. The extracts were purified by chromatography on silica gel using $CHCl_3$ as a developing solvent to give XV (230 mg), which was recrystallized from MeOH as colourless prisms, m.p. 216–219°, $[\alpha]_D^{24}$ −81·5° (*c* = 1·01, $CHCl_3$). (Found: C, 75·22; H, 10·14. Calcd. for $C_{21}H_{34}O_3$: C, 75·40; H, 10·25%). IR $\nu_{max}^{CHCl_3}$ 1694 cm^{-1} (COOH). NMR ($CDCl_3$); 0·94, 1·21, 1·26 (singlets, 3H each $-\overset{|}{\underset{|}{C}}-CH_3$), 3·11 (3H singlet, $-OCH_3$).

Methyl 16α-*methoxy*-(−)-*kauran*-19-*oate* (XVII). A suspension of the finely powdered XV in ether was treated with ethereal diazomethane under ice cooling. The crude methyl ester was purified by chromatography on silica gel using benzene as a solvent to give XVII as colourless scales (from MeOH), 119·5–120·5°, $[\alpha]_D^{24}$ − 81·7° (*c* = 2·01, $CHCl_3$). (Found: C, 75·85; H, 10·36. Calcd. for $C_{22}H_{36}O_3$: C, 75·81; H, 10·41%). IR $\nu_{max}^{CCl_4}$ 1733 cm^{-1} ($COOCH_3$). NMR ($CDCl_3$): 0·82, 1·15, 1·26 (singlets, 3H each, $-\overset{|}{\underset{|}{C}}-CH_3$), 3·12, 3·64 (singlets, 3H each, $-OCH_3$, $-COOCH_3$).

16α-*Methoxy*-(−)-*kauran*-19-*ol* (XVIII). To a stirred suspension of LAH 4 (2·00 g) in anhyd ether (150 ml) was added dropwise a soln of XVII (1·82 g) in ether (100 ml). The mixture was stirred at room temp for 30 min and then refluxed for 1 hr. After working up the usual way, the product was purified by chromatography on silica gel using benzene and $CHCl_3$ as developing solvents. Elution with $CHCl_3$ afforded XVIII (1·67 g), which was recrystallized from MeOH as colourless fine needles, m.p. 138·5–139·5°, $[\alpha]_D^{24}$ −41·5° (*c* = 2·00, $CHCl_3$). (Found: C, 78·34; H, 11·30. Calcd. for $C_{21}H_{36}O_2$: C, 78·69; H, 11·32%). IR $\nu_{max}^{CCl_4}$ 3645 cm^{-1} (OH). NMR ($CDCl_3$): 0·96, 1·02, 1·28 (singlets, 3H each, $-\overset{|}{\underset{|}{C}}-CH_3$), 3·13 (3H singlet, $-OCH_3$),

16α-*Methoxy*-(−)-*kauran*-19-*al* (XVI). To a suspension of CrO_3 (1·2 g) in pyridine (15 ml) was added a solution of XVIII (1·10 g) in pyridine (12 ml). The mixture was stirred for 4 hr under ice cooling. After working up in the usual way, the crude XVI (983 mg) was rapidly recrystallized from a mixture of MeOH (15 ml) and water (2 ml) as colourless silky needles, m.p. 103–107°, $[\alpha]_D^{24}$ − 55·0° (*c* = 1·00, $CHCl_3$). (Found: C, 79·06; H, 10·74. Calcd. for $C_{21}H_{34}O_2$: C, 79·19; H, 10·76%). IR $\nu_{max}^{CCl_4}$ 1723 cm^{-1} (CHO). NMR ($CDCl_3$): 0·88, 0·98, 1·28 (singlets, 3H each, $-\overset{|}{\underset{|}{C}}-CH_3$), 3·13 (3H singlet, $-OCH_3$), 9·78 (1H doublet, *J* = 1·5 Hz, —CHO).

Autoxidation of 16α-*methoxy*-(−)-*kauran*-19-*al* (XVI) (*concentration dependency*). The 0·5 ml of 0·2, 1·0, 5·0 and 25% benzene solns of XVI in small test tubes (6 mm × 45 mm) with stopper were allowed to stand at room temp and the decomposition process of each solutions was followed by TLC as illustrated in Fig 1.

Autoxidation of XVI *in a diluted benzene solution*

Preparation of the peracid XX. A 1% soln of XVI (250 mg) in benzene was allowed to stand at room temp for 6 days. After evaporation of the solvent *in vacuo*, the residue was rapidly recrystallized from a small amount of benzene to yield XX (120 mg) as colourless prismatic needles, m.p. 111·5–112·5° (dec, the crystals crack at 88–89° and become white powders), $[\alpha]_D^{25}$ − 73·55° (*c* = 1·39, $CHCl_3$). (Found: C, 72·23; H, 9·73. Calcd. for $C_{21}H_{34}O_4$: C, 71·96; H, 9·78%). NMR see Table 1.

This peracid (XX) can be prepared alternately by bubbling air into a 5% benzene soln of XVI for 10 hr.

Decomposition of the peracid (XX). On standing at room temp for 7 hr, the decomposition of XX in a crystalline state began to produce XXIII and after 1 week, half the amount of XX was decomposed. The benzene solns of XX at 1·0 and 25% were allowed to stand at room temp, respectively. After 2·5 hr, silky needles deposited from the 25% soln, which were shown to be XXIII with a trace of XXIV by TLC. However XX in the doluted solution (1%) remained unchanged even after a few weeks.

Autoxidation of XVI *in a concentrated benzene solution*

Preparation of the hydroperoxides (XXI *and* XXII) *and hydroxide* (XXIII). A soln of XVI (333 mg) in benzene (1·5 ml) was allowed to stand at room temp for 9 days. During this time, a fairly large amount of crystals were deposited. Evaporation of the solvent *in vacuo* gave a semi-crystalline residue, which was chromatographed on silica gel using benzene and benzene–ether as developing solvents. Elution with benzene–ether (99 : 1) gave a pure sample of XXI (9·0 mg), which was recrystallized from acetone–water (2 : 1) as colourless prismatic needles, m.p. 145·5–147°, $[\alpha]_D^{25}$ − 54·3° (*c* = 0·70, $CHCl_3$). Mass spectrum: M^+ *m/e* 322 corresponding to the molecular formula $C_{20}H_{34}O_3$. IR $\nu_{max}^{CCl_4}$ 3356 cm^{-1} (OOH). NMR see Table 1.

Further elution with benzene–ether (97 : 3) yielded mixtures of XXII and XIX, and XIX and XXIII, each of which was separated to an acidic (XIX) and a non-acidic substance (XXII or XXIII) by shaking with 1% KOH solution and ether.

The acidic XIX (70 mg) was identical with the authentic sample of XV by mixed fusion, TLC, IR and NMR spectra.

Compound XXII (35·5 mg) was recrystallized from aqueous acetone as colourless needles, m.p. 166·5°–167·5°, $[\alpha]_D^{25}$ − 21·87 (*c* = 0·96, $CHCl_3$). Mass spectrum: M^+ *m/e* 322 corresponding to the molecular formula $C_{20}H_{34}O_3$. IR $\nu_{max}^{CCl_4}$ 3556 cm^{-1} (OOH). NMR see Table 1.

Compound XXIII (70 mg) was recrystallized from hexane as colourless fine needles, m.p. 137–138°, $[\alpha]_D^{25}$ − 34·0° (*c* = 1·00, $CHCl_3$). (Found: C, 78·52; H, 11·15. Calcd. for $C_{20}H_{34}O_2$: C, 78·38; H, 11·18%). IR $\nu_{max}^{CCl_4}$ 3610 cm^{-1} (OH). NMR see Table 1.

Stability of the hydroperoxides (XXI *and* XXII). A 10% soln of XXI and a sat soln (*ca* 2·7%) of XXII in benzene were allowed to stand at room temp for 4 days. TLC of each soln indicated that decomposition did not occur.

NaI *Reduction of the hydroperoxide* (XXI). To a soln of XXI (11·6 mg) in abs MeOH (1·0 ml), anhyd ether (0·2 ml) and glacial AcOH (1 drop) was added NaI (60 mg) and allowed to stand at room temp for 4 days. After evaporation of the solvent *in vacuo*, the residue dissolved in ether was washed with $Na_2S_2O_3$ soln and then water. An oily product thus obtained was chromatographed on silica gel using benzene and benzene–ether (97:3) to afford XXIV as colourless crystals, m.p. *ca* 113°, $[\alpha]_D^{24}$ − 57·80° (*c* = 0·35, $CHCl_3$). NMR see Table 1.

NaI *Reduction of the hydroperoxide* (XXII). XXII (6·4 mg) was reduced with NaI (60 mg) as described above. The product, m.p. 135–137°, was identical with the sample of XXIII by TLC, IR and NMR spectra.

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