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# TERPENES OF *PODOCARPUS LAMBERTIUS*

JAYR DE PAIVA CAMPELLO and SEBASTIÃO FERREIRA FONSECA

Instituto de Quimica, Universidade Estadual de Campinas, Campinas, São Paulo, Brazil

and

CHING-JER CHANG and ERNEST WENKERT*

Department of Chemistry, Indiana University, Bloomington, Indiana 47401, U.S.A.

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**Key Word Index**—*Podocarpus lambertius*; Podocarpaceae; bark; leaves; terpenes; structural analysis.

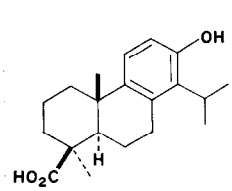
**Abstract**—Sitosterol and the following terpenic compounds have been isolated from the bark of *Podocarpus lambertius*: 3β-hydroxytotarol, 4β-carboxynortotarol, and macrophyllic and lambertic acids. The leaves yielded sitosterol, stigmastan-3β,5α-diol-6-one, isopimaric acid, phyllocladene, isophyllocladene, 8,9-abieten-15-ol and 17-isophyllocladenol.

## INTRODUCTION

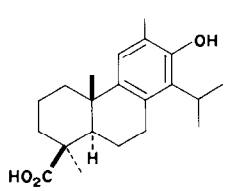
*Podocarpus lambertius*, commonly known as pinheiro bravo [1], is one of two Brazilian species of the genus *Podocarpus* and a tree widely distributed throughout southern Brazil. In continuation of our chemical studies of podocarps [2] an investigation of the terpenic constituents of *P. lambertius* bark and leaves was undertaken.

## RESULTS AND DISCUSSION

Three bark constituents were identified as sitosterol, 4β-carboxynortotarol (**1**) [3–8] and macrophyllic acid (**2**) [7,9,10], while the remaining substances $C_{20}H_{30}O_2$ and $C_{20}H_{28}O_3$ required full structural analysis.




(1)

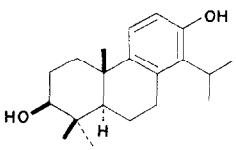



(2)

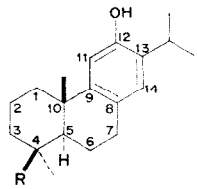
The IR spectrum of the compound $C_{20}H_{30}O_2$ revealed an aromatic ring and hydroxy groups and the facile transformation into a diacetate confirmed the presence of two hydroxy functions. The PMR spectrum of the diol in $CDCl_3$ showed two aromatic one-proton doublets at 6·50 and 6·96 ppm with *J*-values of 9 Hz each, which moved to 7·05 and 7·15 ppm in deuteriopyridine, indicating two vicinal aromatic hydrogens one of which was *ortho* to a phenolic hydroxy group [11]. The presence of an isopropyl group on the benzene ring [1·31 ppm (*d*, 3, *J* 7 Hz, Me), 1·33 (*d*, 3, *J* 7 Hz, Me), 3·28 (*m*, 1, CH)] and its vicinality to the hydroxy function was indicated by the deuteriopyridine shift (1·67—Me, 1·71—Me, 3·50—CH). These observations indicated a totarol skeleton or its biogenetically less likely 11-isopropyl-12-hydroxy isomer. The PMR spectrum exhibited also a one-proton doublet of doublets at 3·28 ppm (*J* 10, 7 Hz), indicative of a *sec*-hydroxy group adjacent to a quaternary carbon center. The high-field position of the oxymethine indicated its equatorial nature [12,13]. The strong deuteriopyridine shift exerted not only on the oxymethine (3·28 → 3·50 ppm) but also on two methyl singlets (0·87 → 1·14, 1·05 → 1·32 ppm) and only a minor shift (Δδ 0·11 ppm) on the remaining methyl singlet suggested a 3β-hydroxytotarol (**3**) relationship. The diol **3** had been reported as a product of borohydride reduction [14] of the naturally occurring 3-totarolone [15] and its MS was recorded [14]. Comparison of the highly characteristic fragmentation pat-

* Present address: Department of Chemistry, Rice University, Houston, Texas 77001, U.S.A.

tern and the other physical properties of an authentic specimen [16] with those of the $C_{20}H_{30}O_2$ product showed the compound to be 3$\beta$-hydroxytotarol (**3**).



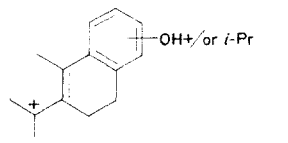
(**3**)



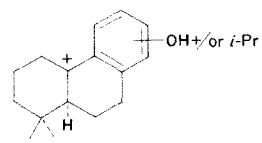
(**4a**) R = H
(**4b**) R = $CO_2H$

The $C_{20}H_{28}O_3$ substance, herewith named lambertic acid, showed IR absorption bands characteristic of hydroxy, carboxy and arene units. Its PMR spectrum in $CDCl_3$ solution revealed two aromatic hydrogen singlets at 6·59 and 6·79 ppm which shifted to 7·21 and 7·08 ppm in deuteriopyridine [11], indicative of a 2,4,5-trialkylated phenol moiety. The presence of an isopropyl group was shown by two methyl doublets at 1·20 (*J* 7 Hz) and 1·22 ppm (*J* 7 Hz) and a methine septet at 3·06 ppm (*J* 7 Hz) and its *ortho* relationship with the phenolic hydroxy group indicated by the deuteriopyridine shift (1·35, 1·36 and 3·64 ppm, respectively). These facts suggested that lambertic acid possessed an environment surrounding the aromatic ring reminiscent of ferruginol (**4**). Besides the methyl groups of the isopropyl function two methyl singlets (1·08 and 1·28 ppm) appeared in the PMR spectrum and were displaced to a single peak at 1·38 ppm in deuteriopyridine. On the basis of a ferruginol skeleton this implies the replacement of one of the C(4) or C(10) methyl groups by a carboxy function and, more specifically, the introduction of a carboxy group spatially proximate to both remaining methyl groups, i.e. a 4$\beta$-carboxy function [17]. The identity of the MS fragmentation pattern of the non-aromatic portion of lambertic acid with that of 4$\beta$-carboxynortotarol (**1**) showed the new compound to be 13-isopropylpodocarpic acid (=4$\beta$-carboxynorferruginol) (**4**) [18].

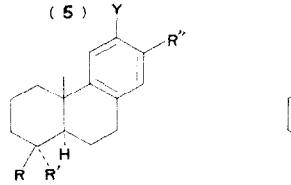
An exhaustive study of the MS fragmentation of tricyclic, aromatic diterpenes of ring A and B low oxidation level has shown that two primary fragmentation patterns involve the production of $M^+$-43 and $M^+$-15 ions (**5** and **6**, respectively) and that the latter undergoes further extensive fragmentation [19, 20]. MS analysis of podocarpic acid (**7a**), dehydroabietic acid (**7b**), **1** and **4b** revealed that the introduction of a carboxyl group at C(4) suppresses the $M^+$-43 peak and produces alternate fragmentation modes of the $M^+$-15 ion, e.g. the formation of ion **8**. Thus the MS indicate the position of the carboxy group. The previously observed enhancement of the intensity of the $M^+$-15 peak (e.g. **6**) in the case of the *p*-hydroxybenzyl cation [19] strikingly differentiated between the aromatic substitution pattern of acids **1** and **4b**.



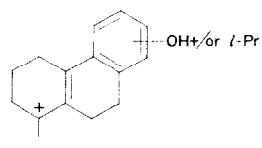

(**5**)




(**6**)



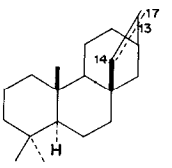
(**7a**) R = $CO_2H$, R′ = Me, R″ = H, Y = OH
(**7b**) R = Me, R′ = $CO_2H$, R″ = *i*-Pr, Y = H



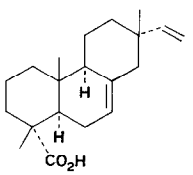

(**8**)

The constituents of the leaves of *P. lambertius* consisted of sitosterol, phyllocladene (**9a**), isophyllocladene (**9b**), isopimaric acid (**10**) and substances $C_{20}H_{34}O$, $C_{20}H_{32}O$ and $C_{20}H_{30}O_3$. The known compounds were identified by comparison with authentic specimens, while the last three compounds necessitated structural determinations.

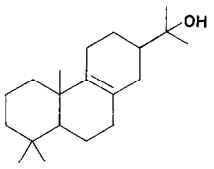
The IR spectrum of the $C_{20}H_{34}O$ compound showed hydroxyl absorption bands and its PMR spectrum five methyl singlets at 0·81, 0·87, 0·89, 1·32 and 1·32 ppm and no signal downfield of 2·4 ppm. The deshielded position of two methyl signals and their deuteriopyridine shift (1·49 ppm) [11] as well as the detection of a fragment of 43·0181 mass units, corresponding to an acetyl ion, in the high-resolution MS revealed the presence of a dimethylcarbinol unit. If it be assumed that the compound is a diterpenic alcohol, its molecular formula and methyl group content requires it to be tricyclic and to contain a tetrasubstituted double bond. Unfortunately the paucity of material left the analysis incomplete and in light of the above data and biogenetic reasoning structure **11** is suggested.



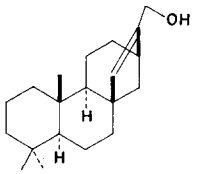

(**9a**) $\Delta^{13(17)}$
(**9b**) $\Delta^{13(14)}$




(**10**)




(**11**)




(**12**)

The $C_{20}H_{32}O$ substance showed IR absorption bands characteristic of hydroxy and olefinic units, which were confirmed by the PMR spectrum. The latter revealed an oxymethylene singlet at 4·17 ppm, which was shifted to 4·43 ppm in deuteriopyridine [11], and an olefinic hydrogen singlet at 5·63 ppm with a deuteriopyridine shift of 5·83 ppm [11]. These facts are accommodated most readily by a 2,3-dialkylallyl alcohol moiety. Three methyl groups appeared at 0·74, 0·81 and 0·85 ppm and were unaffected by deuteriopyridine. Since the molecular formula suggested a tetracyclic configuration, the unusual shielding of one of the methyl groups (0·74 ppm) was reminiscent of the effect of the nuclear double bond of hibaene and isophyllocladene (**9b**) upon their angular methyl groups. Comparison of the chemical shifts of the C(4) and C(10) methyl groups of the alcohol with the methyl shifts of isophyllocladene showed them to be identical. Hence the new alcohol was 17-isophyllocladenol (**12**).

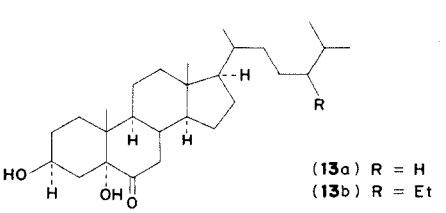
The $C_{29}H_{50}O_3$ product revealed IR absorption bands for hydroxyl and acyclic or six-membered cyclic, saturated keto groups. Its PMR spectrum revealed six methyl signals whose chemical shifts and multiplicities were indicative of a stigmastane skeleton. One of the oxygens reflected a *sec*-hydroxy group in view of a 3·95 ppm oxymethine PMR multiplet which was shifted to 5·03 ppm on acetylation of the product. The remaining presence of a hydroxy absorption band in the IR spectrum of the monoacetate showed the three oxygens of the natural compound to be part of a keto group, of one *sec*-hydroxy function and of a *tert*-hydroxy unit. The ORD spectrum of the $C_{29}H_{50}O_3$ compound exhibited a negative Cotton effect at 323 nm characteristic of 4-, 6-, 7- and 16-ketocholestanes [21]. Base-induced deuteration of the substance showed the presence of two hydrogens $\alpha$ to the carbonyl group. If it be assumed that the compound is an all-*trans* 3-hydroxysteroid, the facts accumulated thus far supported a 3,5-dihydroxy-6-keto or 3,8-dihydroxy-7-keto structure. The 16-keto system was excluded by the carbonyl group not being part of a five-membered ring, while the 4-keto system could be dismissed by the persistence of the 3-oxymethine PMR signal even after deuteration and by the abnormally high field position and far too complex multiplicity for an $\alpha$-ketooxymethine moiety. The two remaining structures could be differentiated by PMR in pyridine [11]. Comparison of the PMR spectra of the natural product in $CDCl_3$ and pyridine revealed no solvent effect on the methyl groups but an extraordinary shift on the methine associated with the *sec*-hydroxy function. The oxymethine multiplet, whose shape (e.g. $W_{1/2} = 16$ Hz) was highly reminiscent of 3$\beta$-hydroxysteroids, shifted 0·73 ppm downfield in pyridine solution. This was in sharp contrast to the $\Delta\delta$ values of 0·25, 0·24, 0·26 and 0·16 ppm for 3$\beta$-hydroxy-5$\alpha$, 3$\beta$-hydroxy-5$\beta$, 3$\alpha$-hydroxy-5$\alpha$ and 3$\alpha$-hydroxy-5$\beta$ steroids, respectively, and reflected the proximity of the *tert*-hydroxy group to the 3$\alpha$-hydrogen and the effect of both hydroxy functions complexed by pyridine acting on this hydrogen. This structural relationship is satisfied only by a 3$\beta$,5$\alpha$-dihydroxy-6-keto system.

In order to confirm the functional groups of the $C_{29}H_{50}O_3$ product and their positions in the steroid framework, the spectral properties of the compound and of its monoacetate were compared with those of 3$\beta$,5$\alpha$-dihydroxy-6-cholestanone (**13a**) [22] and of its acetyl derivative.* The $\delta$ and J

* The relationship of the individual functional groups to their environment was shown also by the products of some simple reactions of the natural substance. While treatment of the latter with $Ac_2O$ and $C_5H_5N$ gave a monoacetate, acetylation under $BF_3$ catalysis yielded a diacetate (m.p. 169–170°). Comparison of the PMR spectra of the two acetates showed that the introduction of the second acetyl group had no effect on any methyl groups but deshielded the 3-oxymethine by 0·23 ppm in conformity with the presence of a 5$\alpha$-hydroxy group oriented 1,3-diaxially toward the 3$\alpha$-hydrogen. Oxidation of the natural product with chromic acid in $C_5H_5N$ gave a hydroxydione (m.p. 251–253°) whose facile, acid-induced dehydration with formation of a conjugated enedione

(*continued*– p. 246)

values of H(3) and H(7) and of the 18-, 19-, 21-, 26- and 27-methyl groups of the natural product and its acetate were nearly identical with those of the corresponding models. This observation suggested that the new plant product is 3β,5α-dihydroxy-6-stigmastanone (**13**b). As a consequence, a partial systhesis from β-sitosterol was undertaken. Treatment of the latter with formic acid and with hydrogen peroxide, followed by hydrolysis with aqueous base gave a triol whose oxidation with N-bromosuccinimide [22] yielded **13**b. The identity of this substance with the natural product fully established its structure.




(**13**a) R = H
(**13**b) R = Et

(**13**)

## EXPERIMENTAL

M.ps. were determined on a Reichert micro hot stage and are uncorrected. ORD measurements were recorded using 1 cm quartz cells. PMR spectra of $CDCl_3$ solutions (unless otherwise noted) with internal TMS were recorded on 60 MHz, 100 MHz and 220 MHz instruments. MS were determined using a direct inlet system. Column chromatography was performed on silica gel (0·05–0·2 mm) and silica gel G and on neutral, activity 1 Merck alumina and TLC on silica gel G or silica GF 254.

*Extraction and isolation. Podocarpus lambertius* Klotzsch was collected in May in the vicinity of Curitiba in the Brazilian state of Paraná. The dried, milled bark (6·9 kg) was extracted ×4 with 6 l. of refluxing $C_6H_6$ each for 24 hr. The extract was evaporated and a hexane solution (1·5 l.) of the residual, brown, viscous oil (89·4 g) extracted under $N_2$ at 20° with ×4 of 250 ml of 5% aq. KOH. Drying of the hexane soln and evaporation yielded 19·6 g of neutral oil. The basic soln was acidified with N HCl to pH 4 and exhaustively extracted with hexane–$Et_2O$ (1:1). Evaporation of the dried extract gave 35·9 g of acidic, partially solid residue.

Chromatography of the neutral oil on silica gel G and elution with hexane–$Et_2O$ (50:1) yielded 744 mg of solid, long-chain fatty alcohols, which were not investigated, and 1·19 g of sitosterol; m.p. 135–137°; identical with an authentic sample. Chromatography of the acidic solid on silica gel G and elution with hexane $Et_2O$ (9:1) yielded 128 mg of 3β-hydroxytotarol (**3**); m.p. 176–178°; $[\alpha]_D^{23}$ + 36·6 (*c* = 0·02, EtOH); UV $\lambda_{max}^{MeOH}$ 281 nm(log ε 3·27); IR (Nujol) OH 3500 (*m*), 3450 (*m*), 3340–3020 (*m*), C=C 1650 (*w*), 1580 (*w*) $cm^{-1}$; PMR δ 0·87, 1·05, 1·17 (*s*, 3, Me each), 1·31, 1·33 (*d*, 3, *J* 7 Hz, *i*-Pr Me each), 2·72 [*ddd*, 1, *J* 18, 12, 6 Hz, H(7α)], 2·97 [*dd*, 1, *J* 18, 7 Hz, H(7β)], 3·28 [*dd*, 1, *J* 10, 7 Hz, H(3α)], 3·28 (*m*, 1, *i*-Pr CH), 6·50 [*d*, 1, *J* 9 Hz, H(12)], 6·96 [*d*, 1, *J* 9 Hz, H(11)]; PMR ($d_5$-pyridine) δ 1·14, 1·28, 1·32 (*s*, 3, Me each), 1·67, 1·71 (*d*, 3, *J* 7 Hz, *i*-Pr Me each), 2·84 [*ddd*, 1, *J* 18, 12, 6 Hz, H(7α)], 3·10 [*dd*, 1, *J* 18, 7 Hz, H(7β)], 3·50 (*m*, 2, OCH, *i*-Pr CH), 7·05 [*d*, 1, *J* 9 Hz, H(11)], 7·15 [*d*, 1, *J* 9 Hz, H(12)]; MS *m/e* 302 ($M^+$), 287 (base) (302 − Me, *m** 272·2), 269 (287 − $H_2O$, *m** 252·2), 243, 227, 215, 201, 175; *m/e* 302·2246 (calc. for $C_{20}H_{30}O_2$: 302·2246). [Diacetate ($Ac_2O/C_5H_5N$, 20°, 48 hr): IR ($CHCl_3$) CO 1750 (*s*), 1730 (*s*) $cm^{-1}$; PMR δ 0·95, 1·23, 1·25 (*s*, 3, Me each), 1·24 (*d*, 6, *J* 7 Hz, *i*-Pr $Me_2$), 2·07, 2·30 (*s*, 3, Ac Me each), 4·82 [*m*, 1, H(3α)], 6·78 [*d*, 1, *J* 9 Hz, H(12)], 7·10 [*d*, 1, *J* 9 Hz, H(11)]; MS *m/e* 386 ($M^+$), 344 (386 − $CH_2CO$, *m** 306·5), 329 (344 − Me, *m** 314·8), 269 (329 − HOAc, *m** 220·5), 175, 43 (base)]. Elution with hexane–$Et_2O$ (1:1) gave 323 mg of 4β-carboxynortotarol (**1**); m.p. 178–180°; $[\alpha]_D^{23}$ + 123·5 (*c* = 0·05, EtOH); UV $\lambda_{max}^{MeOH}$ 281 nm (log ε 3·35); IR (Nujol) OH 3550 (*m*), 3420 (*m*) CO 1705 (*s*), C=C 1580 (*w*) $cm^{-1}$; PMR δ 1·07, 1·31 (*s*, 3, Me each), 1·32 (*d*, 6, *J* 7 Hz, *i*-Pr $Me_2$), 2·58 [*ddd*, 1, *J* 18, 12, 6 Hz, H(7α)], 2·93 [*dd*, 1, *J* 18, 7 Hz, H(7β)], 3·26 (*sept*, 1, *J* 7 Hz, *i*-Pr CH), 6·48 [*d*, 1, *J* 9 Hz, H(12)], 6·96 [*d*, 1, *J* 9 Hz, H(11)]; PMR ($d_5$-pyridine) δ 1·45, 1·48 (*s*, 3, Me each), 1·64, 1·72 (*d*, 3, *J* 7 Hz, *i*-Pr Me each), 2·90 [ddd, 1, *J* 18, 12, 6 Hz, H(7α)], 3·14 [*dd*, 1, *J* 18, 7 Hz, H(7β)], 3·50 (*sept*, 1, *J* 7 Hz, *i*-Pr CH), 7·05 [*d*, 1, *J* 9 Hz, H(11)], 7·15 [*d*, 1, *J* 9 Hz, H(12)]; MS *m/e* 316 ($M^+$), 301 (316 − Me, *m** 286·7), 283 (301 − $H_2O$, *m** 266·1), 255 (301 − $HCO_2H$ *m** 216·0), 213 (255 − $C_3H_6$, *m** 177·9), 43 (base); *m/e* 316·2018 (calc. for $C_{20}H_{28}O_3$: 316·2038). [Acetate ($Ac_2O$/$C_5H_5N$, 20°, 48hr): m.p. 239–242°; IR ($CHCl_3$) OH 3480 (*w*), 3350–2400 (*m*), CO 1755 (*s*), 1690 (*s*), C=C 1580 (*w*) $cm^{-1}$; PMR δ 1·12, 1·31 (*s*, 3, Me each), 1·23, 1·25 (*d*, 3, *J* 7 Hz, *i*-Pr Me each), 2·29 (*s*, 3, Ac Me), 2·66 [*ddd*, 1, *J* 18, 12, 6 Hz, H(7α)], 2·95 [*dd*, 1, *J* 18, 7 Hz, H(7β)], 3·25 (*sept*, 1, *J* 7 Hz, *i*-Pr CH), 6·73 [*d*, 1, *J* 9 Hz, H(12)], 7·14 [*d*, 1, *J* 9 Hz, H(11)]; MS *m/e* 358 ($M^+$), 343, 316 (base) (358 − $CH_2CO$, *m** 278·9), 301 (316 − Me, *m** 286·7), 255 (301 − $H_2O$, *m** 216·0), 213 (255 − $C_3H_6$, *m** 177·9)]. Further elution with hexane–$Et_2O$ (1:1) afforded 206 mg of lambertic acid (**4**b); m.p. 252–254°; $[\alpha]_D^{23}$ + 121·5 (*c* 0·03, EtOH); UV $\lambda_{max}^{MeOH}$ 283 nm (log ε 3·49); IR (Nujol) OH 3360 (*m*), 3250–2500 (*m*), CO 1695, C=C 1620 (*w*), 1580 (*w*) $cm^{-1}$; PMR δ 1·08, 1·28 (*s*, 3, Me each), 1·20, 1·22 (*d*, 3, *J* 7 Hz, *i*-Pr Me each), 2·72 [*m*, 2, $H_2$(7)], 3·06 (*sept*, 1, *J* 7 Hz, *i*-Pr CH), 6·59 [*s*, 1, H(11)], 6·79 [*s*, 1, H(14)]; PMR ($d_5$-pyridine) δ 1·35, 1·36 (*d*, 3, *J* 7 Hz, *i*-Pr Me each), 1·38 (*s*, 6, $Me_2$), 2·92 [*m*, 2, $H_2$(7)], 3·64 (*sept*, 1, *J* 7 Hz, *i*-Pr CH), 7·08 [*s*, 1, H(14)], 7·21 [*s*, 1, H(11)]; MS *m/e* 316 ($M^+$, base), 301 (316-Me, *m** 286·7), 283 (301-$H_2O$, *m** 266·1), 255 (301-$HCO_2H$, *m** 216·0), 213 (255-$C_3H_6$, *m** 177·9); *m/e* 316·1957 (calc. for $C_{20}H_{28}O_3$: 316·2038). [Acetate ($Ac_2O$/$C_5H_5N$, 20°, 72 hr): m.p. 182–184°; IR ($CHCl_3$) OH 3400–2400 (*m*), C=O 1750 (*s*), 1690 (*s*), C=C 1620 (*w*) $cm^{-1}$; PMR δ 1·11, 1·31 (*s*, 3, Me each), 1·15, 1·18 (*d*, 3, *J* 7 Hz, *i*-Pr Me each), 2·13 [*m*, 2, $H_2$(7)], 2·27 (*s*, 3, Ac Me), 2·76 (*m*, 1, *i*-Pr CH), 6·80 [*s*, 1, H(14)], 6·92 [*s*, 1, H(11)]; MS *m/e* 358 ($M^+$), 343, 316 (base) (358-$CH_2CO$, *m** 278·9), 301 (316-Me, *m** 286·7), 255 (301-$H_2O$, *m** 216·0), 213 (255-$C_3H_6$, *m** 117·9)], [Δδ(acetate-phenol) of **4**: H(11) 0·21, H(14) 0·13, *i*-Pr Me, 0·04, 0·05; Δδ (acetate-phenol) of **3** H(11) 0·24, H(14) 0·15, *i*-Pro Me, 0·05, 0·04]. Finally, elution

(*continued from* p. 245)

(m.p. 167–168°) exhibited the β-relationship of the hydroxy group to the newly formed ketone. Comparison of the PMR spectra of the hydroxydione and the natural product showed a Δδ value of 0·22 ppm for the 19-methyl group, similar to 0·18 ppm for Δδ (cholestanone-cholestanol), thus indicative of the change of a 3β-hydroxy group into a keto function. Comparison of the PMR spectra of the hydroxydione in $C_5H_5N$ and $CDCl_3$ solutions [11] revealed strong $C_5H_5N$ shifts on H(4α) [2·91 → 3·21 ppm (*d*, *J* 16 Hz)] and H(7α) [2·77 → 3·06 ppm (*t*, *J* 13 Hz)], characteristic of the effect of a 5α-hydroxy group.

with the same solvent mixture yielded 755 mg of macrophyllic acid (**2**); m.p. 234–236°; identical with an authentic sample.

*P. lambertius* leaves were collected as the bark above, dried and ground in a Wiley mill. The powder (8·9 kg) was extracted in × 7 with 6 l. of refluxing $C_6H_6$ each for 24 hr. The extract was evaporated and a hexane soln (2·5 l.) of the residual, viscous brown oil (380 g) extracted under $N_2$ at 20° with × 5 of 500 ml of 5% aq. KOH. Drying of the hexane soln and evaporation yielded 190·2 g of neutral oil. The alkaline soln was brought to pH 4 with N HCl, saturated with NaCl and exhaustively extracted with hexane–$Et_2O$ (1:1). After drying and evaporation 71·7 g of a semi-solid was obtained.

Chromatography of the neutral oil on alumina and elution with hexane gave 406 mg of a solid, long-chain hydrocarbon, 1·40 g of a solid long-chain fatty alcohol and 147 mg of isophyllocladene (**9b**) [containing a phyllocladene (**9a**) impurity]; m.p. 102–104°; identical with an authentic sample (PMR analysis revealed the presence of 12% **11a**.) Elution with hexane–$Et_2O$ (50:1) gave 26 mg of a long-chain ketone, 3·99 g of sitosterol; mp 135–137°; identical with an authentic specimen; and to 117 mg of an alcohol tentatively assigned structure **11**: m.p. 174–176°; $[\alpha]_D^{23}$ + 4·0° (c 0·01, $CHCl_3$); IR (Nujol) OH 3450–3100 (*w*) $cm^{-1}$; PMR $\delta$ 0·81, 0·87, 0·89 (*s*, 3, Me each), 1·32 (*s*, 6, $Me_2$); PMR ($d_5$-pyridine) $\delta$ 0·80, 0·86, 0·89 (*s*, 3, Me each), 1·49 (*s*, 6, $Me_2$); MS *m/e* 290 ($M^+$), 275 (290-Me, *m** 260·8), 272 (290-$H_2O$, *m** 255·1), 257, 232, 217 (232-Me, *m** 202·9), 41 (base); *m/e* 290·2616 (calc. for $C_{20}H_{34}O$ 290·2610). Elution with hexane–$Et_2O$ (4:1) afforded 34 mg of 17-isophyllocladenol (**12**): m.p. 125–127°; $[\alpha]_D^{20}$ + 23·7° (*c* 0·04, $CHCl_3$); IR (Nujol) OH 3400–3150 (*m*), C=C 1640 (*w*) $cm^{-1}$; PMR $\delta$ 0·74, 0·81, 0·85 (*s*, 3, Me each), 4·17 (*s*, 2, $OCH_2$), 5·63 (*s*, 1, olefinic H); PMR ($d_5$-pyridine) 0·75, 0·80, 0·85 (*s*, 3, Me each), 4·43 (*s*, 2, $OCH_2$), 5·83 (*s*, 1, olefinic H); MS *m/e* 288 ($M^+$, base), 273 (288-Me, *m** 258·8), 270 (288-$H_2O$, *m** 253·1), 257 (288-$CH_3OH$, *m** 229·3), 255 (273-$H_2O$, *m** 238·2), 245; *m/e* 288·2416 (calc for $C_{20}H_{32}O$: 288·2453). Elution with $Et_2O$ gave 1·91 g of long-chain alcohols which were not investigated. Chromatography of the solid, derived from the base extraction, on silica gel and elution with hexane–$Et_2O$ (50:1) yielded 145 mg of isopimaric acid (**10**); m.p. 162–164°; identical with an authentic sample. Elution with hexane–$Et_2O$ (1:1) produced 190 mg of 3$\beta$, 5$\alpha$-dihydroxy-6-stigmastanone (**13b**); m.p. 246–248°; $[\alpha]_D^{22}$ −14·4° (*c* = 0·01, MeOH); ORD (*c* = 0·387, dioxane) $\Phi_{350}$ − 940°, $\Phi_{326}$ − 4250°, $\Phi_{316}$ − 1990°, $\Phi_{309}$ 0°, $\Phi_{286}$ + 4200°, $\Phi_{234}$ + 1217°, $\Phi_{210}$ + 6200°; CD (*c* 0·387, dioxane) $\Delta\delta_{350}$ 0, $\Delta\epsilon_{318}$ − 1·42, $\Delta\epsilon_{308}$ − 1·90, $\Delta\epsilon_{301}$ − 1·65, $\Delta\epsilon_{243}$ − 0·87, $\Delta\epsilon_{215}$ − 0·29; UV $\lambda_{max}^{CHCl_3}$ 301 nm (log $\epsilon$ 1·57); IR (Nujol) OH 3490 (*m*), 3300 (*m*), CO 1710 (*s*) $cm^{-1}$; PMR $\delta$ 0·64, 0·79 (*s*, 3, Me each), 0·81, 0·82 (*d*, 3, *J* 6 Hz, Me each), 0·87 (*t*, 3, *J* 6 Hz, Me), 0·91 (*d*, 1, *J* 7 Hz, Me), 2·09 [*dd*, 1, *J* 13, 5 Hz, H(7$\beta$)], 2·71 [*t*, 1, *J* 13 Hz, H(7$\alpha$)], 3·95 (*m*, 1, $W_{1/2}$ 16 Hz, OCH); PMR ($d_5$-pyridine) $\delta$ 0·65, 0·96 (*s*, 3, Me each), 0·86, 0·89 (*d*, 3, *J* 6 Hz, Me each), 0·90 (*t*, 3, *J* 6 Hz, Me), 0·98 (*d*, 3, *J* 7 Hz, Me), 2·62 [*dd*, 1, *J* 13, 5 Hz, H(7$\beta$)], 3·13 (*t*, 1, *J* 13 Hz, H(7$\alpha$)], 4·68 (*m*, 1, $W_{1/2}$ 16 Hz, OCH); MS *m/e* 446 ($M^+$, base), 428 (446-$H_2O$, *m** 410·7), 413, 410, 400, 385, 375, 371, 359, 346 (446-$C_5H_8O_2$, *m** 267·8), 331 (346-Me, *m** 316·6); *m/e* 446·3723 (calc. for $C_{20}H_{50}O_3$: 446·3760). For comparison PMR of **13a**: 0·63, 0·77 (*s*, 3, Me each), 0·81 (*d*, 6, *J* 6 Hz, $Me_2$), 0·90 (*d*, 3, *J* 7 Hz, Me), 2·05 [*dd*, 1, *J* 13, 5 Hz, H(7$\beta$)], 2·75 [*t*, 1, *J* 13 Hz, H(7$\alpha$)], 3·95 (*m*, 1, $W_{1/2}$ 18 Hz); PMR ($d_5$-pyridine) of **13a**: 0·62, 0·93 (*s*, 3, Me each), 0·88 (*d*, 6, *J* 6 Hz, $Me_2$), 0·94 (*d*, 3, *J* 7 Hz, Me), 2·56 [*dd*, 1, *J* 13, 5 Hz, H(7$\beta$)], 3·13 [*t*, 1, *J* 13 Hz, H(7$\alpha$)], 4·67 (*m*, 1, $W_{1/2}$ 18 Hz, OCH). [Monoacetate of **13b** ($Ac_2O/C_5H_5N$, 20°, 72 hr): m.p. 252–254°; IR ($CHCl_3$) OH 3580 (*w*), 3500–3400 (*w*), CO 1708 (*s*) $cm^{-1}$; PMR $\delta$ 0·63, 0·81 (*s*, 3, Me each), 0·84 (*d*, 6, *J* 6 Hz, $Me_2$), 0·87 (*t*, 3, *J* 6 Hz, Me), 0·92 (*d*, 3, *J* 7 Hz, Me), 1·99 (*s*, 3 Ac Me), 2·75 [*t*, 1, *J* 13 Hz, H(7$\alpha$)], 5·03 (*m*, 1, $W_{1/2}$ 16 Hz, OCH); MS *m/e* 488 ($M^+$), 428 (488-HOAc, *m** 376·1), 410 (428-$H_2O$, *m** 392·7), 385, 375, 367, 359, 346, 331 (346-Me, *m** 316·6). For comparison PMR of monoacetate of **13a**: 0·65, 0·81 (*s*, 3, Me each), 0·86 (*d*, 6, *J* 6 Hz, $Me_2$), 0·91 (*d*, 3, *J* 7 Hz, Me), 2·00 (*s*, 3, Ac Me), 2·76 [*t*, 1, J 12 Hz, H(7$\alpha$)], 5·02 (*m*, 1, $W_{1/2}$ 18 Hz, OCH)].

*Synthesis of 3$\beta$, 5$\alpha$-dihydroxy-6-stigmastanone* (**13b**). A stirred suspension of 2·66 g of sitosterol in 27 ml of 90% $HCO_2H$ was heated at 80° for 12 hr. After cooling, the resultant paste was treated with 3 ml of 30% $H_2O_2$ and shaken at 20° for 12 hr. Boiling $H_2O$ (40 ml) was added and the mixture cooled and filtered. The ppt was dried and dissolved in 80 ml MeOH. A soln of NaOH (3 ml of 20%) was added and the mixture boiled for 10 min, acidified with conc HCl, diluted with 30 ml of $H_2O$ and filtered. The ppt was dried at 120° for 12 hr. (While the crude triol was used in the next reaction, crystallization of a small sample from hexane gave stigmastan-3$\beta$,5$\alpha$,6$\beta$-triol, m.p. 242–244°.) N-Bromosuccinimide (200 mg) was added to a soln of the ppt in 10 ml of dioxane and 2·5 ml of $H_2O$ at 25° and the mixture kept at this temp. for 10 min. It then was cooled in ice, treated with 5 ml of $H_2O$ and filtered. The ppt (700 mg) was washed with 50% MeOH and crystallized from MeOH yielding **13b**; m.p., m.m.p. 246–248°; identical with the natural product.

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