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RESINS OF THE DACRYDIUM AND PODOCARPUS GENERA—THE WOOD RESIN FROM *Dacrydium cupressinum* (RIMU)

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Summary

A brief account is given of the chemistry of the resinous constituents from the woods of a number of species of *Podocarpus* and *Dacrydium*. The neutral portion (0·7 per cent. on the dry wood) of the resin extracted from the wood of *Dacrydium cupressinum* with alcohol is shown to contain the diterpenoids totarol, ferruginol, 9–keto–ferruginol (apparently identical with sugiol), the sesquiterpene macrocarpol, a phytosterol (probably β sitosterol), and a crystalline wax. A structure for totarol is suggested and discussed briefly. This chemical evidence suggests a close relationship of *D. cupressinum* to *Podocarpus*, and absence of any relationship to *Dacrydium*.

INTRODUCTION

Many species of the *Dacrydium* and *Podocarpus* genera of the *Podocarpaceae* produce resinous woods which are noted for their durability and are recognized as valuable timber trees. This is true of a number of New Zealand species, and the resinous constituents of some of them have been examined chemically.

Previous investigations have dealt with the resins extracted with alcohol from the woods of *Dacrydium colensoi*[1], *D. biforme*[2,3], and *D. kirkii*[4]. These resins consist mainly of the neutral oxygenated diterpenes manoöl (I), manoyl oxide (II), and 3–ketomanoyl oxide (III). *D. bidwillii* has also been found to contain manoöl[5]. The correctness of the structures proposed for these compounds was finally confirmed[6] by the establishment of a direct relationship with abietic acid. The manoöl type of diterpenoids possess perhydronaphthalene structures substituted in such a manner that they readily cyclize to perhydrophenanthrenes. On dehydrogenation they yield 1.5,6–trimethylnaphthalene and 1.7,8–trimethylphenanthrene. *D. cupressinum*, in contrast to the Dacrydium species mentioned above, contains a different type of diterpenoid, namely, the phenol, podocarpic acid* (IV)[7,8,9] present as crystalline heart-shake resin. It is shown below that three further phenolic diterpenoids are present in *D. cupressinum*. These phenolic diterpenoids, which appear to be characteristic of the woods of *Podocarpus* species, possess a substituted perhydrophenanthrene structure, and on dehydrogenation yield substituted phenanthrene phenols and hydrocarbons.

Podocarpus dacrydioides and the Javanese pine, *P. cupressinum*, both contain podocarpic acid as heart-shake resin[7,10].

P. ferrugineus yields an oleo-resin, the main constituent of which is the non-crystalline phenolic diterpenoid ferruginol (V)[9,11]. On dehydro-

*Podocarpic acid, which has the formula $C_{17}H_{26}O_3$, may be regarded as derived from a diterpene which has lost an isopropyl group.

genation, ferruginol yields 6-retenol (VI) and pimanthrene (VII), while the hydrocarbon ferruginane (VIII), obtained on catalytic hydrogenation, yields only retene (IX) on dehydrogenation.

P. totara wood, upon alcohol extraction, yields mainly the phenolic diterpenoid totarol[12,13], possibly (X), which has been dehydrogenated to 1-methyl-7-hydroxyphenanthrene (XI) and 1-methylphenanthrene (XII). Catalytic hydrogenation of totarol yields the hydrocarbon totarane (XIII?), which dehydrogenates to 1-methyl 8-isopropylphenanthrene (XIV)[14,15]. From the wood of *P. totara* there has also been isolated [16], a crystalline phenolic diterpene alcohol ($C_{20}H_{30}O_2$) of melting-point 230 to 231°, apparently hydroxy totarol, and a further phenol, $C_{30}H_{44}O_2$, of melting-point 222 to 223°, probably a triterpenoid.

P. spicatus contains a heart-shake resin, the main constituents of which are matai-resinol and conidendrin. The established structures of these compounds show them to be lignans[17,18,19]. The solvent extractives of the wood of this species have not yet been examined.

THEORETICAL

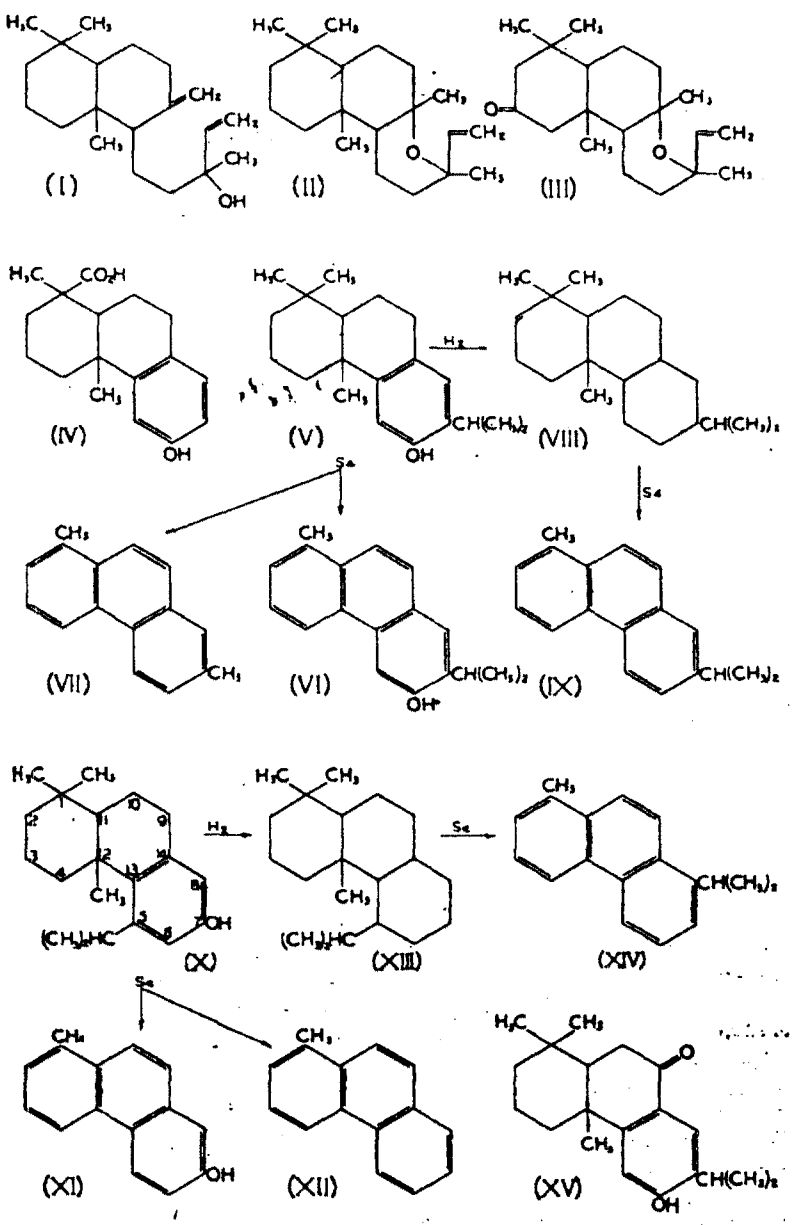
This paper reports the composition of the neutral portion of the resin extracted from the wood of *Dacrydium cupressinum* with alcohol. Unlike other species of the genus referred to above, this wood contains only a small proportion of neutral material (0·7 per cent. of the dry wood), and no compounds of the manoöl (I) type could be isolated from it. The acidic portion of the resin (6 per cent. of the dry wood) contains, among other acids, a considerable amount of podocarpic acid[20] (IV).

The neutral part of the resin after distillation, acetylation, and chromatographing yielded the acetates of ferruginol (V) and totarol to the extent of 5 per cent. and 10 per cent. respectively, together with about 1 per cent. each of 9-ketoferruginol (XV), a phytosterol probably β-sitosterol, and a crystalline wax. The saturated sesquiterpene alcohol macrocarpol, first isolated from *Cupressus macrocarpa*[21], was obtained from the low-boiling unacetylated fractions. The high proportion of neutral material unaccounted for consisted of resinous residues after distillation and chromatography. They probably contain higher terpenoids or highly oxygenated diterpenes.

Totarol was found to be phenolic, a fact which has not been reported previously, although the presence of three double bonds had been demonstrated[14], consequently the identity of the product under investigation was not realized until it had been dehydrogenated to 1-methyl-7-hydroxyphenanthrene. The substance was then identified as totarol by mixed melting-point with an authentic sample. Its phenolic nature was indicated by coupling with diazotized *p*-nitroaniline and by a slight but definite solubility in sodium hydroxide solution. This was confirmed from the absorption spectra of the acetate and of the free phenol and the shift in wavelength maximum of the latter in alkaline solution.

In the tentative structure (X) now suggested for totarol, one quaternary methyl group at C_1 and one at C_{12} are so placed by analogy with all other polycyclic diterpenoids of known structure. If the isopropyl group is placed at C_8, as suggested by the dehydrogenation of totarane to (XIV), the resulting structure would not conform to the isoprene rule and would be the first known exception among the diterpenoids.

Although instability of the substituents of the aromatic ring on dehydrogenation is evident in both totarol and ferruginol, the two reactions are not parallel. Thus in ferruginol both substituents remain intact or the isopropyl group is partially eliminated with simultaneous loss of the hydroxyl group. In totarol, the isopropyl group is entirely eliminated, while the hydroxyl group may or may not be lost.



C_8 and C_9 are the only positions in the molecule where the isopropyl group could be placed in order to produce a structure which complies with the isoprene rule. If placed at C_9, the dehydrogenation of totarol would be expected to follow a similar pattern to that of ferruginol. It has been shown[22] that an alkyl group at C_8 is unstable on dehydrogenation. Thus 1,5-dimethyl-5,6,7,8-tetrahydrophenanthrene yields mainly 1,8-dimethylphenanthrene and a small amount only of the 1,5 isomer on selenium dehydrogenation. It has been suggested previously[14] that group migration must occur in order to explain the chemistry of totarol. Apparently movement of the isopropyl group to C_8 during hydrogenation or subsequent dehydrogenation was implied.

Ferruginol was isolated as the acetate and identified by mixed melting-point.

9-Ketoferruginol was isolated as colourless needles. The determination of the structure of this substance and comparison with sugiol from *Cryptomeria japonica*[23,24], with which it is probably identical, will appear in a separate note.

Macrocarpol and the 3,5-dinitrobenzoate were identified by mixed melting-points with authentic specimens. The reported composition and absence of double bonds in macrocarpol were confirmed. A crystalline acetate (m.p., 43 to 44°) and benzoate (m.p., 76°) were prepared for the first time.

The phytosterol, $C_{29}H_{50}O$, had m.p. 139 to 140° and $[\alpha]_D^{18°}$ −30°. These physical properties and those of the acetate agree with those recorded for β-sitosterol.

The colourless *crystalline wax* of m.p. 63 to 64° was saturated and had the composition of $C_{24}H_{48}O_2$ or $C_{25}H_{50}O_2$, but was not investigated further.

It will be apparent from the introductory part of this paper that a clear distinction exists between the types of extractives obtained from the woods of the *Dacrydium* and *Podocarpus* species. *Dacrydium cupressinum*, however, is seen to be an exception, showing a close chemical relationship to *Podocarpus* in that it contains only phenolic diterpenoids characteristic of that genus.

Biological tests indicate that totarol, ferruginol, and 9-ketoferruginol are without oestrogenic activity.

EXPERIMENTAL

Rimu sawdust was extracted with alcohol, and the concentrate, after addition of water, extracted with ether. The ether soluble resin was separated into neutral resin (0·7 per cent. of the dry wood) and resin acids (6 per cent. of the dry wood) by extraction with sodium carbonate solution.

The dark amber-coloured neutral resin when distilled gave the following fractions: Frs. 1–3, b.p. 160 to 200°/5 mm., amber oils (4 g.); Frs. 4–9, b.p. 150 to 210°/0·1 mm., amber resins (21 g.); Fr. 10, b.p. 240 to 260°/3·5 mm., dark resin (9 g.); residue (17 g.). All fractions were mixtures which could not be readily crystallized or chromatographed. Separation was effected by acetylation of the fractions and chromatographing on alumina in petroleum ether solution.

Totaryl acetate: Fractions 4–9 were combined after acetylation with acetic anhydride and pyridine and chromatographed on alumina; the benzene eluate gave a white crystalline solid which was recrystallized several times from alcohol as prisms, m.p. 125°, $[\alpha]_D^{20°}$ + 45° in alcohol, λ max. 266 mμ, log ε max. 2·7 in alcohol. A mixed melting-point with an authentic specimen of totaryl acetate was not depressed (Short and Stromberg[14] gave melting-point of totarol acetate 122° $[\alpha]_D^{18°}$ + 44·6° in ether).

Totarol: Totaryl acetate (100 mg.) was refluxed with N-alcoholic potassium hydroxide solution for 30 minutes. The product (85 mg.) extracted with ether from the reaction mixture in the usual manner was recrystallized twice from light petroleum, prisms, m.p. 128 to 129°. A mixed melting point with totarol, m.p. 132°, was not depressed. A pure sample of totarol in alcohol gave λ max. 280 mμ, log ε max. 3·3, and in 0·1 N-alcoholic potassium hydroxide solution the absorption spectrum showed a wavelength shift to λ max. 288 mμ, log ε max. 3·3.

Totaryl benzoate: Totarol (30 mg.) from *P. totara*, pyridine (2 ml.), and benzoyl chloride (0·3 ml.) were mixed and stood for several hours. Recrystallization of the product three times from alcohol gave *totaryl benzoate*, m.p. 146 to 147°.

Found	— — — —	C, 83·0; H, 9·0 %
Calculated for $C_{27}H_{34}O_2$	— —	C, 83·0; H, 8·8 %

Totarol from rimu was benzoylated in the same way and the product was recrystallized twice from alcohol, plates, m.p. 147 to 148°. A mixed melting-point of the two benzoates was not depressed. *1-methyl-7-hydroxyphenanthrene benzoate*: Totaryl acetate (280 mg.) was dehydrogenated with selenium (500 mg.) at 300 to 320° for 22 hours. The reaction mixture was extracted with ether and the product, after recrystallization several times from benzene, gave 1-methyl-7-hydroxyphenanthrene (60 mg.), m.p. 197 to 198°. It formed an unstable picrate, m.p. 163 to 164°. 1-methyl-7-hydroxyphenanthrene (20 mg.) was benzoylated and the product was recrystallized from alcohol (21 mg.). Recrystallization twice from acetone gave *1-methyl-7-hydroxyphenanthrene benzoate*, m.p. 203 to 204°

Found	— — — — —	C, 84·3; H, 5·4 %
Calculated for $C_{22}H_{16}O_2$	— —	C, 84·1; H, 5·8 %

Ferruginyl acetate: The light petroleum eluate obtained when the acetate of fractions 4–9 were chromatographed gave a crystalline solid (2·6 g.), which was recrystallized several times from alcohol, m.p. 82°, λ max. 269 mμ, log ε max. 3·3 (in alcohol). A mixed melting-point with authentic ferruginyl acetate, m.p. 82°, was not depressed.

Ferruginol: Ferruginyl acetate (1 g.) was refluxed in 5 per cent. alcoholic potassium hydroxide solution for 40 minutes. The product (900 mg.) separated from light petroleum as a white powder, m.p. 57 to 59°, λ max. 283 mμ, log ε max. 3·5 (in alcohol).

Ferruginyl benzoate: Ferruginol (90 mg.) was benzoylated and the product (90 mg.) was recrystallized from light petroleum, m.p. 155°, not depressed by an authentic sample of ferruginyl benzoate, m.p. 157 to 158°.

9–Ketoferruginol: When both acetylated and unacetylated fractions 4–9 were chromatographed, 9–ketoferruginol separated from the acetone eluate as colourless needles (total yield, 550 mg.). It was crystallized several times from benzene as needles, m.p. 295 to 297°, $[\alpha]_D^{20°}$ + 20° (in alcohol); λ max. 233, 285 mμ, log ε max. 4·2, 4·1.

Found — — — — —	C, 79·6; H, 9·4 %
Calculated for $C_{20}H_{28}O_2$ — —	C, 80·0; H, 9·4 %

9–Ketoferruginyl methyl ether: 9–Ketoferruginol (35 mg.), dimethyl sulphate (2 ml.), potassium carbonate (2 g.), and acetone (10 ml.) were refluxed together for two hours. The product was crystallized from aqueous alcohol as plates (8 mg.), m.p. 136 to 137°.

9–Ketoferruginyl benzoate: 9–Ketoferruginol (20 mg.) was benzoylated and the product purified by sublimation (16 mg.) and recrystallized twice from alcohol, needles, m.p. 185 to 186°.

Found — — — — —	C, 80·2; H, 8·1 %
Calculated for $C_{27}H_{32}O_3$ — —	C, 80·2; H, 8·0 %

9–Ketoferruginol semicarbazone: 9–Ketoferruginol (20 mg.), semicarbazide hydrochloride (25 mg.), and sodium acetate (40 mg.) were dissolved in alcohol (2 ml.) and heated on the waterbath for three hours. The product was largely unchanged material and was heated again with similar quantities of reagents for 24 hours. The product was extracted with hot benzene from unchanged material and recrystallized from alcohol as pale yellow prisms, m.p. 242 to 245°.

Macrocarpol: Fractions 1–3 were chromatographed and the benzene eluate (400 mg.) was sublimed twice (100–110°/0·2 mm.) and crystallized from light petroleum as prisms (200 mg.), melting-point 111 to 112°, $[\alpha]_D^{18°}$ + 15° (alcohol, c = 1).

Found — — — — —	C, 80·9; H, 11·8 %
Calculated for $C_{15}H_{24}O$ — —	C, 81·0; H, 11·8 %

The compound gave no coloration with tetranitromethane and was recovered unchanged after shaking in ethyl acetate solution with hydrogen and platinum oxide catalyst for two hours. A mixed melting-point with macrocarpol, m.p. 111 to 112°, from *Cupressus macrocarpa*[21] was not depressed.

Macrocarpyl acetate: Macrocarpol (40 mg.) was acetylated with pyridine and acetic anhydride. The oily product (38 mg.) crystallized on standing, and purification by chromatography and sublimation under reduced pressure gave *macrocarpyl acetate*, m.p. 43 to 44°.

Found — — — — —	C, 77·2; H, 10·8 %
Calculated for $C_{17}H_{28}O_2$ — —	C, 77·2; H, 10·7 %

Macrocarpyl benzoate: Macrocarpol (35 mg.) was benzoylated and the product was chromatographed. The light petroleum eluate by crystallization from alcohol and sublimation under reduced pressure gave *macrocarpyl benzoate* (22 mg.), m.p. 76°.

Found — — — — —	C, 80·7; H, 9·4 %
Calculated for $C_{22}H_{30}O_2$ — —	C, 80·9; H, 9·3 %

Macrocarpyl 3:5–dinitrobenzoate: Macrocarpol (20 mg.) and 3:5–

dinitrobenzoyl chloride (100 mg.) were dissolved in benzene (1 ml.), pyridine (1 ml.) was added, and the mixture stood for several hours. The product (40 mg.) was recrystallized from alcohol as needles (25 mg.), m.p. 158 to 159°. A mixed melting-point with an authentic specimen, m.p. 158 to 159°, from *Cupressus macrocarpa* was not depressed.

Phytosteryl acetate: Fraction 10 was acetylated and chromatographed. The fraction eluted slowly with light petroleum was rechromatographed and crystallized from alcohol (yield 350 mg.), m.p. 120 to 122°. Recrystallization several times from alcohol gave the acetate as needles, melting-point 124 to 125°, $[\alpha]_D^{20°}$ — 35° (chloroform, c = 2).

Found — — — — —	C, 81·4; H, 11·4 %
Calculated for $C_{31}H_{52}O_2$ — —	C, 81·5; H, 11·5 %

Phytosterol: The acetate (30 mg.) was refluxed in N-alcoholic potassium hydroxide solution (4 ml.) for 30 minutes. The solution was diluted with water and the product filtered off. Recrystallization twice from alcohol gave the phytosterol as plates (14 mg.), melting-point 139 to 140°, $[\alpha]_D^{18·5°}$ — 30° (chloroform, c = 1). The Liebermann Burchard reaction gave a purple colour.

A waxy compound in the first petrol ether eluates was obtained on chromatographing fractions 4–9 after acetylation. Recrystallization several times from alcohol to constant melting-point gave colourless plates, m.p. 63 to 64° (400 mg.).

Found — — —	C, 78·2; H, 13·1 %; M (Rast), 384
Calculated for $C_{21}H_{48}O_2$	C, 78·2; H, 13·1 %; M, 368
Calculated for $C_{25}H_{50}O_2$	C, 78·5; H, 13·2 %; M, 382

The compound gave no coloration with tetranitromethane.

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