PODOCARPACEAE

SOME CONSTITUENTS OF PODOCARPUS SALIGNA*

M. SILVA† and M. HOENEISEN
Departamento de Botanica, Universidad de Concepcion, Chile

and

P. G. SAMMES
Chemistry Department, Imperial College, London, SW7

(Received 12 May 1971)

BECAUSE of the current interest in Podocarpus species14 and our project dealing with Chilean flora,5 a preliminary examination of Podocarpus saligna D. Don, a plant indigenous to Chile, has been made. The leaves were collected August 1968, 15 km north of Concepcion. After cold extraction with ethanol, the soluble material was re-extracted in turn with benzene, ethyl acetate, and n-butanol. From the benzene extract was isolated the saturated hydrocarbon fraction, the major component being nonacosane, and also β-sitosterol, identified by conversion into its acetate. Fern-9(11)-ene was isolated from the ethyl acetate fraction together with some of its isomer, probably isofernene. A lactone, ν\textsubscript{max} 1770, 1735 and 1710 cm\(^{-1}\) was also isolated from this fraction. Its mass spectrum showed a molecular weight of 376, corresponding to C\(_{20}H_{34}O_7\), and it showed a base peak at m/e 289, due to loss of 87 mass units and analysed as loss of C\(_4\)H\(_2\)O\(_2\). Its NMR spectrum was reminiscent of those from the highly oxygenated norditerpenes such as the nagilactones,6 podolactones,7 and imumakilaactones,8 isolated from similar species. On the basis of extensive proton decoupling experiments9 the lactone has been assigned structure (I). Mass spectral fragmentation occurs preferentially to form the ion a.

* Partly supported by the Organization of American States (Grant PMC-8/1)
† To whom enquiries should be made

5 M SILVA, G PORLETI and P G SAMMES, Phytochem In press.
9 Carried out at Imperial College. We thank Mr P N Jenkins for his help with these experiments, the full details of which will be published elsewhere.
A further lactone of related, but as yet undefined constitution has also been isolated in small amounts from this extract. Cyanidin and delphinidin were isolated from the leaves after extraction and which are therefore present originally as leucoanthocyanidins. The leaves did not show any insect moulting hormone activity.

**EXPERIMENTAL**

Instruments used were as described previously. Material for chromatography was supplied by Merck. The dried, ground leaves (10 kg) were extracted to exhaustion with EtOH at room temp. After removal of solvent in vacuo the residue (2.9 kg) was treated with H₂O and the precipitate (0.8 g) discarded. The solution was extracted with benzene, EtOAc and n-BuOH. The solvents were removed in vacuo at <45 °C to yield the fraction from benzene (16.7 g), EtOAc (160 g), and n-BuOH (820 g).

The benzene fraction (16 g) was chromatographed through alumina (grade III, 200 g) to yield the following compounds:

**Hydrocarbon fraction** 25 mg eluted with light petroleum. After recrystallization from MeOH had mp 57–60 °C. Mass spectral analysis showed the principal component to be nonacosane. This fraction was not further investigated.

**β-Sitosterol** 40 mg eluted with benzene–light petroleum, m p (EtOH) 132–135 °C, un-depressed by mixed m p with authentic material. Acetylation with Ac₂O in pyridine at room temp afforded the acetate, m p 120–122 °C.

A portion (48 g) of the EtOAc fraction was chromatographed through silica gel (loading ratio 1:200) to give the following compounds:

**Hydrocarbon fraction** 3.4 g eluted with benzene, m p 50–70 °C. TLC on silica gel G–10 % AgNO₃, using cyclohexane as solvent, indicated two major components. The first was identical to authentic fern-9(11)-ene (R, 0.61) and the second component of the same Rₜ as isofernene (R, 0.65). Direct spectral comparison (IR) of the former with an authentic sample confirmed the assignment.

**Lactone A** 370 mg eluted with 1:1 EtOAc–benzene, m p (EtOAc) 259–260 °C, [α]D₂O +40.3 ° (c 0.77) pyridine), λmax 2900, 2866, 1770, 1735 and 1710 cm⁻¹. NMR bands (CDCl₃ + 1 drop D₂O-pyridine) 8 76 (3Hs, Me₃), 8 75 (3Hd, J 7 Hz, Me₃), 8 69 (1Hd, J 5 3 Hz, 5-H) 8 1–8 5 (2Hm, 7a-H, 7b-H), 7 48–7 85 (2Hm, 8-H), 6 92 (1Hd, J 19 Hz, 1-H), 6 70 (1Hm, 2-H), 6 40 (3Hs, MeO), 5 77 (1Hdd, J 2 4, 11 5 Hz, 14-H), 5 14 (1Hm, 6-H), 4 32 (1Hd, J 2 5 Hz, 11-H). Mass spectral bands at m/z 376 (M⁺, 0.8 %, corresponding to C₂₀H₂₄O₇), 358 (0.1), 361 (0.1), 346 (1.2), 332 (1.0), 317 (0.2), 289 (100), 274 (2.0), 260 (7), 245 (10), 105 (10), 91 (10) (Found C, 61.53, H, 6.05. C₂₀H₂₄O₇ requires C, 60.90, H, 6.65 %). After attempted acetylation (Ac₂O/pyridine) starting material was recovered. NaBH₄ reduction in EtOH at room temp gave a complex mixture. The material was stable to brief treatment with dill HCl but reacted after a prolonged period.

**Lactone B** 35 mg eluted by EtOAc, m p 280–300 °C, contaminated by traces of lactone A. The structure of this lactone is under investigation. TLC investigation of the n-BuOH extract showed no further major components.

**Detection of leucoanthocyanidins** The powdered, dried leaves (200 g) were refluxed in 2 N HCl for 40 min. The acid was extracted with amyl alcohol and chromatographed on Whatman paper No 3 using both the Forestal and BAW solvent systems. The Rₜ values for the two compounds detected corresponded to cyanidin and delphinidin.

Acknowledgements—We thank the Fund for Overseas Research Grants and Education, New York, for financial support. We also acknowledge with gratitude the help of Prof. C. Marticorena with the botanical determination of plant material.

---

10 We thank Dr D. H. S. Horn for carrying out the tests for insect moulting hormone activity.
12 Carried out at Imperial College with samples provided by Dr G. Mellows.
13 E. C. Bate-Smith and N. H. Lerner, Biochem J 58, 126 (1953).

**Key Word Index**—Podocarpus saligna, Podocarpaceae, terpenes, fern-9(11)-ene, norditerpene, leucoanthocyanidins.