

THE VOLATILE OILS OF THE GENUS *EUCALYPTUS* (FAMILY MYRTACEAE)*

V.† THE LEAF OIL OF *E. AGGREGATA* DEANE & MAIDEN

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Eucalyptus aggregata Deane & Maiden is placed by Blakely¹ in his Section Macrantherae, Series Subexsertae, and is a well-defined species found on damp sites at elevations of from about 500 m to about 1300 m on the central and southern highlands of New South Wales. An isolated and very small occurrence has been described by Carolan² from Woodend in Victoria, separated by some 270 miles from the southernmost extent hitherto recorded for this species in New South Wales, but these trees cannot be distinguished, either morphologically or chemically, from those of the larger northern populations.

The oil of *E. aggregata* has been examined by Baker and Smith,^{3a} who identified (+)- α -pinene and two pentyl esters: pentyl phenylacetate in material from Rydal, N.S.W., and pentyl eudesmate in material from the vicinity of Braidwood, N.S.W. Their original specimen of eudesmic acid, isolated from the saponification of its ester, was shown by Birch and Elliot⁴ to be 3,4,5-trimethoxybenzoic acid. However, it is clear from the data of Baker and Smith that they failed to recognize that the oil from this species is heavier than water; evidently most of their product was lost during steam distillation, and hence the major component of the oil was not identified by them.

The results of this investigation show that the oil is predominantly (c. 90%) a single ester, β -phenylethyl phenylacetate. Lower-boiling fractions, amounting to approximately 1% of the oil, were found to consist chiefly of (+)- α -pinene, (–)- and (\pm)-limonene, and *p*-cymene. The remainder was made up largely of *n*-pentyl phenylacetate, methyl 3,4,5-trimethoxybenzoate, and a new ester, β -phenylethyl 3,4,5-trimethoxybenzoate. Free β -phenylethanol occurred, as well as free phenylacetic acid and 3,4,5-trimethoxybenzoic acid. The last two, obtained in the routine search for alkali-soluble components of the oil, may of course have been hydrolysis products. A waxy solid, apparently of hydrocarbon nature, and found in the highest-boiling fraction, may have been derived from the leaf-wax.

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¹ Blakely, W. F., "A Key to the Eucalypts." 2nd Edn, p. 141. (Forestry & Timber Bureau: Canberra 1955.)

² Carolan, Patricia M., *Victorian Nat.*, 1964, **81**, 117.

³ Baker R. T., and Smith, H. G., "A Research on the Eucalypts and their Essential Oils." 2nd Edn, (a) pp. 318–19, (b) pp. 115–16, 403–4. (Government Printer: Sydney 1920.)

⁴ Birch, A. J., and Elliot, Patricia, *J. chem. Soc.*, 1953, 355.

The Tasmanian *E. rodwayi* R. T. Bak. & H. G. Sm. was included by Blakely in *E. aggregata*, but a comparison of the chemical data for the leaf oils found both by Baker and Smith^{3b} and by the present authors supports the opinion of Johnson⁵ that they are two distinct species.

Experimental

All melting points are uncorrected. Light petroleum had b.p. 40–60°. Vacuum distillations were carried out in a Podbielniak Mini-Cal 3475 fractionating column using Heli-grid packing. Gas chromatography was conducted in a Bodenseewerk Perkin-Elmer Vapor Fractometer 116, using $\frac{1}{8}$ in. by 6 ft aluminium columns packed with polyethylene glycol adipate on Celite for compounds of b.p. <100° at 9.5 mm, and Apiezon L on Celite for compounds of b.p. >100° at 10 mm. Infrared spectra were obtained from a Perkin-Elmer Infracord 137 spectrophotometer. Microanalyses were carried out by the Australian Microanalytical Service, Melbourne.

(a) Isolation of Volatile Material

Fresh foliage was steam distilled in a tin-lined apparatus to yield mobile oils denser than water; ether extractions of the aqueous distillate yielded further quantities of oil. Material was collected at Rydal, N.S.W. (on the same collection site of Baker and Smith), at Hartley Vale, N.S.W., and at Woodend, Vic., and crude oils having the following characteristics were obtained: oil yield, 0.13 to 0.33%; d_{4}^{25} 1.0625 to 1.0866; n_D^{20} 1.5346 to 1.5392; α_D +1.50° to +2.28°; acid no., 1.3 to 1.7; ester no., 208.5 to 218.0. Rydal (foliage from a single tree): oil yield, 0.23%; d_{4}^{25} 1.0730; n_D^{20} 1.5375; α_D +3.80°; acid no., 2.0; ester no., 214.0. Woodend: oil yield, 1.06% (high yield due to distillation in all-glass cohobation apparatus); d_{4}^{25} 1.0232; n_D^{20} 1.5200; α_D -0.36°.

E. rodwayi foliage from various Tasmanian collections yielded oils having the following characteristics: d_{4}^{25} 0.9061 to 0.9203; n_D^{20} 1.4626 to 1.4951; α_D -7.75° to -10.15°; acid no., 0.85 to 1.7; ester no., 5.3 to 9.0; cineole content (*o*-cresol method) 63 to 70%.

Bulked crude oils (110 ml) of *E. aggregata* were fractionally distilled at a pressure of 9.5 mm.

(b) Identification of Constituents

(i) (+)- *α* -Pinene.—A fraction of the oil (0.65 ml), b.p. 35–65° at 9.5 mm, was submitted to gas chromatography, and a homogeneous fraction corresponding to a peak with the same retention time as *α* -pinene was condensed and found to give an infrared spectrum identical with that of an authentic specimen of *α* -pinene. *α* -Pinene fractions obtained from the various oils had α_D up to +39.2°.

(ii) (\pm)-*Limonene* and (-)-*limonene*.—Gas chromatography of a fraction (0.65 ml), b.p. 35–65° at 9.5 mm, showed the presence of limonene by retention time. A homogeneous fraction collected from the column had an infrared spectrum identical with that of an authentic specimen. Bromination of the fraction yielded the dibromide of (\pm)-limonene, m.p. and mixed m.p. 123–124°. From the Rydal oil a dibromide, m.p. 104–105°, was obtained from a terpene fraction having α_D -9.2°.

(iii) *p*-Cymene.—Gas chromatography of a fraction (0.3 ml), b.p. 65–90° at 9.5 mm, using a picric acid-methoxypolyethylene glycol column, separated a fraction from the monoterpenes. This fraction was collected and found to give an infrared spectrum identical with that of an authentic specimen of *p*-cymene.

(iv) β -Phenylethanol.—A fraction (0.28 ml), b.p. 95–110° at 9.5 mm, was shown, by a comparison of its retention time during gas chromatography with that of an authentic specimen, to contain β -phenylethanol as its main component. The infrared spectrum of a homogeneous gas chromatography fraction was identical with that of authentic material.

(v) *n*-Pentyl phenylacetate.—A fraction (1 ml), b.p. 128–130°/9.5 mm, was found to be mainly *n*-pentyl phenylacetate by a comparison of the retention time and infrared spectrum of a homo-

⁵ Johnson, L. A. S., *Contr. N.S.W. nat. Herb.*, 1962, **3**, 105.

geneous gas chromatography fraction with those of an authentic synthetic specimen. The synthetic ester had b.p. $268^{\circ}/763$ mm; d_4^{20} 0.9821; n_D^{20} 1.4864.

(vi) *Methyl 3,4,5-trimethoxybenzoate*.—A fraction (1 ml), b.p. 166° at 9.5 mm, solidified on cooling, and on recrystallization from light petroleum yielded a white crystalline solid, m.p. $82-83^{\circ}$, which was identified as methyl 3,4,5-trimethoxybenzoic acid by a mixed melting point determination and a comparison of the infrared spectrum with that of an authentic synthetic specimen.

(vii) *β -Phenylethyl phenylacetate*.—That portion of the oil, b.p. $>166^{\circ}/9.5$ mm (89 g), on recrystallization from ethanol yielded as the main product β -phenylethyl phenylacetate, m.p. 29° , identified by a mixed melting point determination and comparison of the infrared spectrum with that of an authentic synthetic specimen. Saponification of the solid ester isolated from the residue yielded phenylacetic acid, m.p. and mixed m.p. 76° , and β -phenylethanol, identified by comparison of the infrared spectrum with that of an authentic specimen.

(viii) *β -Phenylethyl 3,4,5-trimethoxybenzoate*.—The ethanolic filtrate of the distillation residue deposited crystals which on recrystallization from ethanol were identified as β -phenylethyl 3,4,5-trimethoxybenzoate, m.p. 60° , by a mixed m.p. determination and comparison of the infrared spectrum with that of an authentic specimen of the synthetic ester (Found: C, 68.25; H, 6.4; O, 25.3. Calc. for $C_{18}H_{20}O_5$: C, 68.3; H, 6.4; O, 25.3%).

(ix) *Unidentified hydrocarbon*.—The distillation residue also contained a small amount of waxy material which was insoluble in light petroleum. Recrystallization from methanol yielded a white waxy solid, m.p. $58-60^{\circ}$ (Found: C, 85.4; H, 14.6%; mol. wt. (Rast), 558).

(x) *Phenylacetic acid*.—Extraction of the crude oil with 10% Na_2CO_3 solution yielded a solid which on recrystallization from water gave phenylacetic acid, m.p. and mixed m.p. 76° .

(xi) *3,4,5-Trimethoxybenzoic acid (eudesmic acid)*.—Extraction of the crude oil with 2% cold NaOH solution yielded a solid which on recrystallization from benzene gave 3,4,5-trimethoxybenzoic acid (0.75%), m.p. and mixed m.p. 169° .

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