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THE STEAM VOLATILE LEAF OILS OF SOME SPECIES OF *EUCALYPTUS* SUBSERIES *STRICTINAE*

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Key Word Index—*Eucalyptus* subseries *Strictinae*; Myrtaceae; volatile leaf oils; mono- and sesquiterpenoids; chemosystematic aspects.

Abstract—The volatile leaf oils of *Eucalyptus stenostoma, E. fraxinoides, E. triflora, E. dendromorpha, E. burgessiana, E. rupicola, E. approximans* subsp. *approximans, E. approximans* subsp. *codonocarpa* and *E. paliformis* were qualitatively very similar to each other but significantly different from the leaf oils of the remaining species of the subseries. Their leaf oils were characterized by high concentrations of *p*-cymene and by the presence of α- and β-phellandrene, variable amounts of piperitone, *trans*- and *cis*-piperitol and *trans*- and *cis*-*p*-menth-2-en-1-ol. The three last named alcohols have not been previously reported from the Myrtaceae.

INTRODUCTION

Some years ago Johnson and Blaxell described several new species of *Eucalyptus* [1, 2] and placed them within the newly proposed subseries *Strictinae* (series *Obliquae*, section *Renantheria*, subgenus *Monocalyptus* [3]. Since some of the species of this subseries exhibit great morphological similarities, an investigation of their volatile oils appeared desirable. Essential oil composition can be, at times, a useful adjunct in the differentiation of botanically close or superficially similar species of *Eucalyptus*, e.g. the pairs *E. maculata*/*E. citriodora, E. smithii*/*E. macarthurii, E. nova-anglica*/*E. cinerea* subsp. *cinerea* [4, and unpublished results, Museum of Applied Arts and Sciences] and more recently, *E. ovata*/*E. brookeriana* [5]. Furthermore, apart from some very early work [6] on the leaf oils of *E. fraxinoides, E. apiculata, E. stricta*, and a brief note on the presence of only the merest trace of volatile oil in the leaves of *E. obtusiflora* [4], the subseries has not been recently investigated. The taxonomy of the subseries as proposed by Pryor and Johnson [3] and previous essential oil data are summarized in Table 1.

RESULTS AND DISCUSSION

A GC/MS investigation of the freshly steam-distilled oils has allowed us to separate a group of nine species and subspecies exhibiting a remarkable qualitative similarity of leaf oil composition (Table 2). However, the unnamed subspecies of *E. approximans*, coded MAKIKB in Pryor and Johnson's system was unavailable in sufficient quantity for a chemical investigation. The remaining four taxa, *E. obtusiflora, E. stricta* and both subspecies of *E. apiculata* yielded qualitatively different and rather more complex oils which will be discussed in a separate communication.

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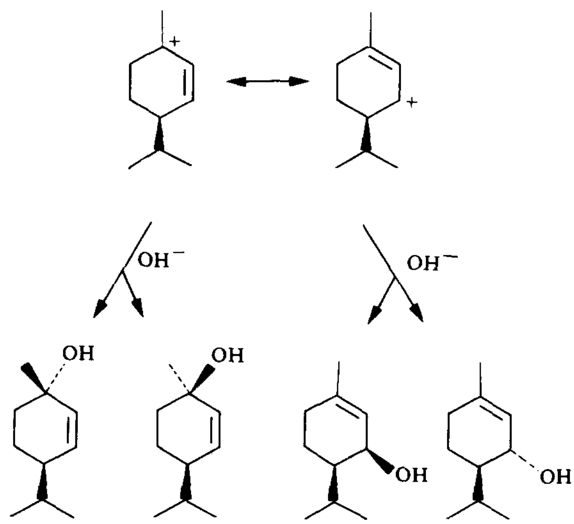
The essential oils of all species listed in Table 2 are characterized by uncommonly high *p*-cymene contents as well as by the presence of three unusual monoterpenoid alcohols *cis*-*p*-menth-2-en-1-ol **1**, *trans*-*p*-menth-2-en-1-ol **2** and *cis*-piperitol **3**. Even though these three alcohols have been found to occur as minor components in several essential oils, e.g. *Zingiber cassumunar* [7], *Mentha piperita* [8, 9], *Chamaecyparis obtusa* [10], dillweed (*Anethum graveolens*) [11], laurel leaf (*Laurus nobilis*) [12], cardamom (*Elettaria cardamomum*) [13], this is, to our knowledge, the first report of their presence, often in relatively large amounts, in the family Myrtaceae. Since *trans*-piperitol **4** co-occurs with **1–3** in all oils examined by us, it is tempting to speculate that they arise in the plant by non-specific hydroxylation of the same allylic carbonium ion (Scheme 1).



1 **2** **3** **4**

Scheme 1.

Table 1. Subseries *Strictinae** of the genus *Eucalyptus*

Superspecies	Species	Subspecies	Pryor and Johnson code	Essential oil
	E. stenostoma L. Johnson et D. Blaxell		MAKIA	
	E. fraxinoides Deane et Maiden		MAKIB	Pinene, citral (?), α-phellandrene, cineole, eudesmol, piperitone
	E. triflora (Maiden) Blakely		MAKIC	
Stricta	*E. dendromorpha* (Blakely) L. Johnson et D. Blaxell		MAKID	
	E. obtusiflora DC.		MAKIE	Trace of oil only
	E. burgessiana L. Johnson et D. Blaxell		MAKIF	
	E. stricta Sieber ex Spreng.		MAKIG	Cineole, α-pinene, aldehydes, eudesmol
	E. apiculata R. T. Baker et H. G. Smith		(MAKIH)	
		apiculata	MAKIHA	Piperitone, cineole, α-pinene, sesquiterpenes
		—	MAKIHB	
	E. rupicola L. Johnson et D. Blaxell		MAKIJ	
	E. approximans Maiden		(MAKIK)	
		approximans	MAKIKA	
		—	MAKIKB	
		codonocarpa Blakely et McKie) L. Johnson et D. Blaxell	MAKIKC	
	E. paliformis L. Johnson et D. Blaxell		MAKIM	

*Classification according to Pryor and Johnson [3].

The relative paucity of reports of natural occurrences of **1–3** may be due to their instability, in particular their ease of dehydration to α-phellandrene, rather than to their rarity. *Cis*-piperitol is known to dehydrate spontaneously at room temperature [14]. Schenck *et al.* [15] reported the decomposition of piperitol during GC using a column packed with a PEG-coated support. In our hands FFAP-coated SCOT columns did not cause any decomposition of either the piperitols or the two isomeric *p*-menth-2-en-1-ols.

Our results indicate that, within the subseries *Strictinae*, leaf oil composition is of limited use as a tool to botanical classification. Whilst we did not observe any qualitative differences amongst the oils of individual trees of the same species, it is also well known that large intraspecific quantitative variation is not uncommon in *Eucalyptus* (e.g. *E. dives* [16], *E. punctata* [17], *E. brookeriana* [5]). Thus, a choice between taxa may be made only where substantial qualitative differences exist. So, for instance, *E. rupicola* and *E. stricta*, which may occur in close proximity on sandstone ridges in the Blue Mountains west of Sydney, may be differentiated with some confidence on essential oil composition alone. Similarly, oil yield and oil composition of *E. dendromorpha*, previously considered to be merely a variety of *E. obtusiflora*, are sufficiently different to support its elevation to species status. The taxonomic position of *E. stenostoma*, though apparently supported on leaf oil composition, is disputed by Brooker [18] who separates it from the *Strictinae* on the grounds of differing floral morphology and places it in a separate monotypic subseries ("*Stenostominae*").

Table 2. GC percentage compositions of the leaf oils of the *Eucalyptus* subseries *Strictinae**

Components	*E. stenostoma* 72–161	*E. fraxinoides* 72–190	*E. triflora* 72–058	*E. dendromorpha* 72–016	*E. burgessiana* 72–020	*E. rupicola* 72–021	*E. approximans* subsp. *approximans* 72–107	*E. approximans* subsp. *codonocarpa* 72–097	*E. paliformis* L.A.S.J. & D.F.B. 490, 4, 1971 (NSW)
α-Pinene	1.7	1.8	1.1	1.0	6.0	14.1	0.4	tr	0.8
β-Pinene	tr	—	tr	0.1	0.1	0.4	0.1	tr	tr
α-Phellandrene	5.6	2.9	2.4	13.4	21.2	2.6	6.0	4.6	1.2
α-Terpinene	2.0	0.4	0.3	3.8	4.3	tr	2.0	0.6	0.2
Limonene	0.3	0.1	tr	0.2	0.2	tr	—	tr	tr
1, 8-Cineole	8.5	1.3	tr	tr	0.1	tr	2.5	6.4	3.6
β-Phellandrene	7.5	13.4	11.5	15.3	16.6	12.3	7.0	5.3	5.5
p-Cymene	25.9	32.7	38.7	24,3	20.7	33.8	26.5	29.8	31.2
Terpinolene	tr	—	0.1	1.0	—	1.6	0.7	0.2	0.3
Un	—	—	tr	0.1	—	0.2	tr	tr	0.8
Un	0.1	0.4	1.0	0.2	0.1	0.3	0.2	—	0.1
trans-p-Menth-2-en-1-ol	16.7	9.7	9.2	6.5	7.6	2.6	10.8	5.4	11.5
Terpinen-4-ol	4.5	1.9	2.6	1.3	3.7	0.7	1.3	0.2	1.4
cis-p-Menth-2-en-1-ol	12.1	7.1	6.7	3.7	4.8	0.4	6.7	3.3	7.5
cis-Piperitol	3.2	2.3	2.2	1.1	1.4	1.4	1.8	0.3	4.3
α-Terpineol	1.7	0.9	0.6	0.6	0.6	0.5	0.3	tr	0.8
Verbenone	tr	—	tr	0.2	—	1.5	0.5	tr	0.4
Piperitone	0.2	0.1	15.7	21.3	0.1	21.1	27.2	38.5	25.7
trans-Piperitol	5.8	4.0	2.9	2.0	3.0	1.3	4.0	2.5	2.4
Myrtenol	tr	—	tr	tr	—	tr	tr	tr	0.8
Un	0.3	0.4	0.2	0.6	—	0.7	0.2	tr	tr
Un	0.3	0.3	0.8	1.2	0.2	2.0	1.6	2.6	1.6
Viridiflorol	—	tr	tr	0.1	0.8	—	tr	—	—
Un	0.4	0.4	4.2	1.0	tr	—	—	tr	tr
γ-Eudesmol	0.2	6.0	tr	—	2.0	—	—	—	—
α-Eudesmol	1.1	5.5	tr	tr	1.9	—	—	tr	tr
β-Eudesmol	1.8	8.5	tr	tr	2.5	—	—	tr	tr

Key: tr—$<0.1\%$; Un—unidentified component. GC column used—FFAP-coated SCOT.
*Herbarium numbers are given for each species.

EXPERIMENTAL

Collection of plant material and isolation of volatile oils. Samples of fresh foliage (400 g/tree) collected from different sides of individual trees were stream-distilled with cohobation for 7 hr in an all-glass apparatus [19]. Yields and physical constants of the oils obtained are shown in Table 3. Herbarium voucher specimen numbers starting with 72-, 74- or 76- refer to specimens held at the Biological and Chemical Research Institute Herbarium. Numbers combining letters and numerals refer to specimens held at the National Herbarium, Royal Botanic Gardens, Sydney.

Identification of oil constituents. Analytical GLC was conducted on a Perkin–Elmer 900 gas chromatograph using 15 m × 0.5 mm i.d. FFAP, SE-30 or Castorwax coated SCOT columns with He as carrier gas. A Hewlett–Packard 3370A. Integrator was used to determine % compositions. Individual components were identified by their retention times and by co-injection with authentic compounds. Further identifications were carried out using GC/MS as described previously [20]. The following major components were isolated and their identity confirmed separately: α-pinene, β-phellandrene, 1, 8-cineole, *p*-cymene, piperitone (IR, n_D^{20}); α-phellandrene (IR of maleic anhydride adduct); *cis-p*-menth-2-en-1-ol, *trans-p*-menth-2-en-1-ol (IR; authentic specimens were prepared from α-phellandrene according to the method of ref. [15])

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Table 3. Yields and physical constants of the leaf oils of the *Eucalyptus* subseries *Strictinae*

Species	Herbarium voucher	Oil yield	n_D^{20}	α_D^{20}	$d_{20°/4°}$
E. stenostoma	D.J.H. NSW 119425, 7.7.1965 (NSW); 72–161	1.7–2.4	1.4807–1.4820	−20.9° to −25.6°	0.8858–0.8996
E. fraxinoides	72–190	2.8	1.4869	−15.1°	0.8994
E. triflora	72–058; 72–060	0.7–0.8	1.4834–1.4856	−23.8° to −25.0°	0.8959–0.9013
E. dendromorpha	72–016; 72–092; 74–030; 74–031	0.3–2.1	1.4834–1.4856	−9.0° to −31.0°	0.8990–0.9256
E. obtusiflora	72–035; 72–038 72–078	0–0.1	—	—	—
E. burgessiana	L.A.S.J. & C.B. NSW 132522 11.3.1953 (NSW); 72–020	1.1–1.5	1.4816–1.4819	−29.4° to −37.0°	0.8782–0.8871
E. stricta	72–015; 72–024; 72–025; 72–135	0.2–0.6	1.4661–1.4877	−3.2° to +17.6°	0.9338–0.9518
E. rupicola	L.A.S.J. NSW 26906, 16.4.1953 (NSW); 72–021; 72–022	1.9–2.8	1.4824–1.4836	−23.5° to −30.4°	0.8959–0.9057
E. approximans subsp. *approximans*	72–107	1.8	1.4827	−21.4°	0.9134
E. approximans subsp. *codonocarpa*	72–097	2.1	1.4831	−23.5°	0.9167
E. paliformis	L.A.S.J. & D.F.B. 490, 4.1971 (NSW)	2.9	1.4825	−18.4°	0.9161
E. apiculata subsp. *apiculata*	72–192; 76–024 76–025; 76–026	0.6–2.2	1.4799–1.4848	−27.0° to +1.6°	0.9006–0.9245
E. apiculata un-named subsp. MAKIHB	72–191; 72–193; 76–035; 76–037 76–038; 76–039; 76–040; 76–041	0.5–1.3	1.4757–1.4857	−2.4° to −12.8°	0.9165–0.9325

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