Phytochemistry, 1964, Vol. 3, pp. 27 to 32. Pergamon Press Ltd. Printed in England

EUDESMOL ISOMERS FROM *CORDIA TRICHOTOMA* WOOD

MARGARET K. SEIKEL and JOHN W. ROWE

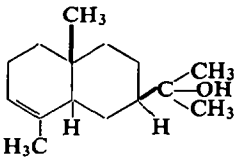
Forest Products Laboratory,[1] Forest Service, U.S. Department of Agriculture

(*Received* 27 *April* 1963)

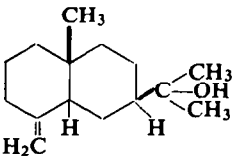
Abstract—White crystals derived from *Cordia trichotoma* wood during manufacture have been identified as a mixture of approximately 48 per cent α-, 35 per cent β-, and 13 per cent γ-eudesmol plus a trace of guaiol. Separation of the four compounds is possible by a combination of gas and preparative thin-layer chromatography.

DISCUSSION

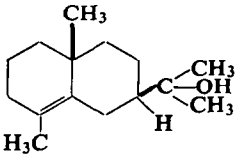
WHEN veneer cut from the South American wood Peteribí, *Cordia trichotoma* (Vell.) Arrab. (Boraginaceae), is stacked and dried,[2] the unexposed surfaces become covered with feathery white crystals. The possible identity of these as eudesmol isomers was suggested by: (a) bands in the infrared spectrum at 3020, 1640, 883, and 799 cm^{-1} for exocyclic methylene and trisubstituted unsaturation; (b) a spectrum typical of a terpene alcohol; and (c) melting point and volatility similar to sesquiterpenes. Final identification of this sample, hereafter referred to as eudesmol-I, as a mixture of α-, β-, and γ-eudesmol with a trace of guaiol was accomplished by comparison with samples of eudesmol mixtures from *Eucalyptus macarthuri* Deane and Maiden[3] (eudesmol-II) and from *Neocallitropsis araucarioides* (R. H. Compton) Florin (= *Callitropsis araucarioides* R. H. Compton)[4,5] of the Cupressaceae (eudesmol-III and -IV) by means of thin-layer and gas chromatography and infrared spectra of the separated α- and β-isomers.



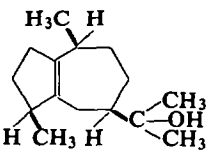
α-Eudesmol



β-Eudesmol



γ-Eudesmol



Guaiol

Preliminary examination by thin-layer chromatography showed that eudesmol-I was a mixture of at least two very similar compounds. Clean separation of these using a method of

[1] Maintained at Madison, Wis., in cooperation with the University of Wisconsin.
[2] The exact conditions of drying are unknown.
[3] F. J. MCQUILLIN and J. D. PARRACK, *J. Chem. Soc.*, 2973 (1956).
[4] R. B. BATES and E. K. HENDRIKSON, *Chem. & Ind.* (London), 1759 (1962).
[5] E. VON RUDLOFF, *Chem. & Ind.* (London), 743 (1962).

overrunning the plates led on a larger scale to the isolation of mg amounts of α- and β-eudesmol, sufficient for infrared analysis. After a single separation, the β-isomer was practically pure, only a trace of material presumed to be guaiol remaining. The α-isomer contained traces of β and was unresolved from γ, which runs the same or slightly faster under the

TABLE 1. CHARACTERISTIC BANDS IN THE INFRARED SPECTRA OF THE EUDESMOL ISOMERS*†

α-Eudesmol cm^{-1}	β-Eudesmol cm^{-1}	γ-Eudesmol cm^{-1}
—	793‡ (w)	—
797 (m)	—	—
—	—	816 (m)
845 (m) § 852 (m) §	—	845 (w)
—	854 (m)	—
—	884 (s)	—
984 (w) § 996 (w) §	984 (m)	multiple shoulders
— ‖	—	—
— ‖	1048 (m)	—
1120 (m)	—	1057 (w)
1142 (s)	—	—
—	1135 (s)	1138 (s)
1200 (m) § 1219 (m) §	—	1150 (s) 1198 (w)
—	1217 (m)	(sh)
1440 (sh)	1260 (s)	—
1450 (s)	1440 (s)	—
1465 (sh)	1450 (sh)	1455 equal or
1630 (w, b)	1470 (s)	1465 fused
	1640 (sp)	(b)

* The many other bands for the three isomers are essentially similar or weak. For figures see reference 3 and K. R. VARMA, T. C. JAIN and S. C. BHATTACHARYYA, *Tetrahedron* **18**, 979 (1962).

† Abbreviations: s = strong, w = weak, m = medium strength, sh = shoulder, sp = sharp, b = broad.

‡ This band is apparently not the characteristic band of α-eudesmol at 797 cm^{-1} since its relative intensity was unchanged after purification of the β-isomer by a second banding of the material.

§ These pairs of bands were of equal intensity for the purest sample of the α-isomer (from eudesmol-II). For the material from eudesmols-I and -III, containing more γ-isomer, the 852 cm^{-1} band was merely a shoulder and the 996 and 1200 cm^{-1} bands were weaker than the 984 and 1219 cm^{-1} bands, respectively.

‖ Absolutely no absorption present in this region.

conditions used. Rebanding should complete the separation of the β-isomer from guaiol and the mixed α- and γ-isomers from the β-isomer. This method of separation can be coupled with gas chromatography, which, although it does not differentiate the α- and β-isomers, readily separates them from the γ-isomer and guaiol.[4] This enables a complete separation to be made of the four sesquiterpene alcohols in these eudesmol mixtures.

Application of separation by thin-layer chromatography to eudesmols-I, -II, and -III served to establish their qualitative identity with each other better than determination of physical constants of the original mixtures. Similar results on the plates and superimposable infrared spectra of the separated fractions were obtained (Table 1). Guaiol, a possible biogenetic relative of the eudesmols, was detected only by gas chromatography, an analytical method which confirmed the similarity of eudesmols-I, -II, and -III (Table 2).

TABLE 2. ANALYSES OF CRUDE SAMPLES OF EUDESMOL AND THE SEPARATED ISOMERS

Sample	Isomer	Approximate percentage by I.R. spectra*	Percentage by gas chromatography
Eudesmol-I	α	45	{83
	β	32	
	γ	9	13
	guaiol (?)†	—	3
Eudesmol-II	α	47	{89
	β	35	
	γ	8	10
	guaiol	—	1
Eudesmol-III	α	31	{64
	β	26	
	γ	15	27
	guaiol	—	8
Eudesmol-IV	α	61	
	β	35	
	γ	—	
α-Isomer from eudesmol-I	α	90	
	β	2	
	γ	11	
α-Isomer from eudesmol-II	α	92‡	
	β	1	
	γ	7	
α-Isomer from eudesmol-III	α	78	{62
	β	1	
	γ	13	38
β-Isomer from eudesmol-III	guaiol	—	1·6

* The γ-eudesmol band is very weak and broad so percentages of this isomer are indicative only.

† The peak was identified in this manner by Bates because of its similarity to the guaiol peak given by eudesmol-III.

‡ Obtained by difference and used as standard for α-isomer.

An estimate of the relative amounts of α- and β-eudesmol in the samples, made from the absorbances of characteristic peaks in the infrared spectra, showed that, in all cases, the α-isomer was predominant. Thin-layer chromatography had suggested the reverse, but this result may be a function of the detecting agent. The γ-isomer was only qualitatively detected (over 5 per cent) by this method of analysis.

That the eudesmols exist as such in the *C. trichotoma* wood was shown by chromatography of a methylene chloride extract of an authentic sample of wood. The typical spots of α- and β-eudesmol resulted. A much lower concentration seemed to be present in the closely related species *C. alliodora* (R. & P.) Cham. None of the eudesmol isomers nor any terpenes have

been previously reported to occur in any *Cordia* species. Eudesmols, however, have been isolated from a variety of different wood oils, especially from the perfumery oils of Eucalyptus species.[6] Just recently, they have been found in *Taxodium distichum* heartwood.[6a]

EXPERIMENTAL

Techniques

Except for γ-eudesmol, all infrared spectra were done with potassium bromide pellets on a double beam infrared recording spectrophotometer, Baird, model AB-1.

The thin-layer chromatograms were run on alumina plates with layers 0·25 mm thick, or for banding 0·50 mm, and developed with benzene-petroleum ether (1:1). To increase the separation of the principal α- and β-eudesmol spots, the inverted plates were developed by a descending method in which the solvent was allowed to run off the plate.[7] The chromatograms were allowed to run 7–18 hr, depending on the thickness of the layer. Spots were visualized by fuming with iodine vapors. For preparative work, streaks were applied to the plates,[9] and the positions of the resulting bands were determined by cross-streaking with dilute permanganate.

To isolate the separated α- and β-isomers, the absorbent containing the desired band was removed from the plates and chromatographically eluted with methylene chloride in a micro-setup;[7,8] 0·5–1·0 mg was eluted completely by 0·2 ml. The solvent was evaporated, preferably by a stream of nitrogen, and the crystalline or oily residue used at once for spectral analysis. After determination of the infrared spectra, the samples were recovered from the powdered potassium bromide pellets by the same method of elution. One sample of β-eudesmol was rebanded and taken completely through the process a second time. Spectral analysis showed 40–45 per cent recovery.

Samples

A crude mixture of eudesmols from *Cordia trichotoma*, eudesmol-I, was obtained from Instituto de Pesquisas Tecnologicas, Estado de Sao Paulo, Sao Paulo, Brazil, both in the form of separated crystals and as a felted mat of fluffy white crystals still adhering to the surface of the veneer. This material melted at 75–75·5° (lit.,[3] 80–83°). Sublimation *in vacuo* (water pump) separated the material from wood scrapings, but the melting point was slightly lowered and broadened, 73–75·5° with considerable preliminary sintering; $[\alpha]_D^{22}$ +35° ($C = 1{\cdot}305$ in $CHCl_3$) (lit.,[3] +31° to +38°).

A mixture of eudesmols from *Eucalyptus macarthuri*, eudesmol-II, furnished by McQuillin,[3] melted at 69–72°. A mixed melting point of eudesmols-I and -II was 71–76°.

A mixture of eudesmols, eudesmol-III, in the form of a partially solidified mass, and liquid γ-eudesmol, 90 per cent pure, from *Neocallitropsis araucarioides* were furnished by Bates.[4] A resublimed sample, eudesmol-IV, from the same source was furnished by von Rudloff.[5]

[6] A. R. PENFOLD, *The Eucalypts; Botany, Cultivation, Chemistry, and Utilization*. Interscience Publishers, New York, 1961.

[6a] H. L. HERGERT, Abstracts of Papers, 9D, Division of Cellulose, Wood, and Fiber Chemistry, 144th Meeting, ACS, Los Angeles, Calif,. April 3, 1963.

[7] M. K. SEIKEL, M. A. MILLET and J. F. SAEMAN, to be published.

[8] M. A. MILLETT, W. E. MOORE and J. F. SAEMAN, to be published.

[9] By an adaptation of the streaking apparatus of S. W. MCKIBBINS, J. F. HARRIS and J. F. SAEMAN, *J. Chromatog.* **5**, 207 (1961).

Results of Thin-Layer Chromatography

The four samples of eudesmol mixtures gave similar but not identical chromatographic patterns. All showed the faster running spot of the α-isomer and the more prominent spot of the β-isomer. The ratios of distance to the front of the α-spot to that of the β-spot were 1·23, 1·22, and 1·25 for eudesmols-I, -II, and -IV. This ratio remained essentially constant even when the distance traveled doubled with a tenfold increase in loading. The ratio for eudesmol-III was consistently higher, averaging 1·34. The relatively high percentage of γ-eudesmol in this material is believed to account for this discrepancy since the α- and γ-isomers are not separated by this procedure. Eudesmol-III also showed two weak, slower running spots with distance ratios compared to the β-isomer of about 0·6 and 0·7.[10] Eudesmols-II and -III showed ultraviolet fluorescent material remaining at the origin; since these samples also showed carbonyl absorption in their infrared spectra, these spots probably indicate autoxidation products. Eudesmol-I, which had been freshly removed from the wood, and recently purified eudesmol-IV were free of these impurities.

By banding on the thin-layer plates, 9 mg of eudesmol-I, 6 mg of eudesmol-II, and 10 mg of eudesmol-III were fractionated, each on two 20 × 20 cm plates with 0·50, 0·25, and 0·50 mm layers, respectively. The α/β distance ratios were respectively 1·27, 1·18, and 1·19. In the third case the very front of the band (ratio 1·31) gave a weaker test and was eluted separately. On spectral analysis the trace of material obtained from it seemed to be much richer in γ-eudesmol than was the heavier contiguous band of the impure α-isomer.

The isolated α-eudesmol was obtained as opaque white solids from eudesmols-I and -II but as a mixture of oil and crystals with the odor of γ-eudesmol from eudesmol-III. The isolated β-eudesmol was in all cases a mixture of oil and crystals. All samples were preserved under nitrogen at less than 0°.

Results of Infrared Spectral Analyses

The infrared spectra of eudesmols-I, -II, -III, and -IV were very similar in respect to wavelength of bands or shoulders,[11] but relative intensities of some bands differed. The three samples of β-eudesmol gave infrared spectra identical in the wavelength and relative intensity of all major and minor bands, with no evidence of more than traces of α-eudesmol. The α-isomers from eudesmols-I and -III were likewise similar, both showing a weak but definite γ-eudesmol band. The α-isomer from eudesmol-II gave a very clearcut spectrum, with peaks at the same wavelengths but in several cases different relative intensities (Table 1); not more than a trace of the γ-eudesmol band was visible.

Approximate percentages of α-, β-, and γ-eudesmol in various samples (Table 2) were determined by calculation from the absorbances of the characteristic bands; 798 cm^{-1} for α-eudesmol, 884 cm^{-1} for β-eudesmol, and 816 cm^{-1} for γ-eudesmol. The 1377 cm^{-1} band (C-methyl) was used as the internal standard. Standard absorbances were obtained from the spectrum of the twice banded β-isomer, the spectrum of the γ-isomer (shown to be 90 per cent pure by gas chromatography), and the spectrum of the α-isomer isolated from eudesmol-II and considered to be approximately 92 per cent pure after corrections for 1 per cent β-eudesmol and a trace of γ-eudesmol (5–8 per cent).

[10] VON RUDLOFF has indicated the presence of α-cadinol in eudesmol-IV.[5] Its presence might explain either the extra spots or other minor differences in our data.

[11] For example, 85 per cent of 46 bands in the spectrum of the eudesmol-I checked within ±0·02 μ with those given by eudesmol-II, a better check than was obtained with 39 bands on two successive spectra of eudesmol-I.

Results of Gas Chromatographic Analysis

Eudesmol-I, -II, and -III and the separated α- and β-isomers isolated from eudesmol-III by thin-layer chromatography were subjected to gas chromatographic analysis for us by Dr. R. B. Bates, University of Illinois (see Table 2 for analytical results). The column was 20 per cent Carbowax on acid-base washed 30/60 firebrick, and it was run at 220° at 36 lb/in^2. The relative retention values in respect to γ-eudesmol were: (a) For eudesmol-III, guaiol 0·75, α- and β-eudesmol 1·21; for eudesmol-II, guaiol 0·76, α- and β-eudesmol 1·23; for eudesmol-I, guaiol (?) 0·74, α- and β-eudesmol 1·21. The β-eudesmol was shown to contain traces of guaiol while the γ-eudesmol impurity in the α-sample was clearly evident.

Acknowledgements—We gratefully acknowledge the gift of samples of eudesmol mixtures from Drs. F. J. McQuillin, University of Durham, R. B. Bates, University of Illinois, and E. von Rudloff, Prairie Regional Laboratory, Canada. In addition, the gas chromatographic analysis was done for us by Dr. Bates. The original material, eudesmol-I, was furnished by Mr. Armondo Russo, Sao Paulo.