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DIFFERENTIATING MANOOL AND 13-EPIMANOOL WITH NMR CHIRAL SHIFT REAGENTS

ANTHONY H. CONNER and JOHN W. ROWE
Forest Products Laboratory,* Forest Service, U.S. Department of Agriculture, Madison, WI 53705, U.S.A.

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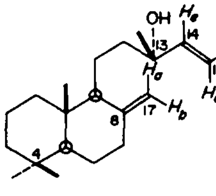
Key Word Index—*Pinus contorta*; *Tsuga heterophylla*; *Pinus* spp.; tall oil; Coniferae chemotaxonomy; NMR chiral shift reagents; diterpene alcohols; manool; 13-epimanool.

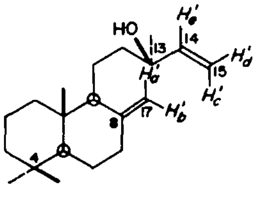
Abstract—Use of NMR chiral shift reagents showed that a compound isolated from *Pinus contorta* bark and *Tsuga heterophylla* wood was 13-epimanool. Southern pine (*Pinus* spp.) tall oil contained a mixture of manool and 13-epimanool.

INTRODUCTION

Manool [8(17),14-labdadien-13-ol]† (**1**) and 13-epimanool [8(17),14-labdadien-13α-ol] (**2**) are tree extractives of interest to the perfumery industry [1,2] and for chemotaxonomic purposes [3]. The compounds are reported to occur in Pinaceae [4–19], Cupressaceae [20–23], Podocarpaceae [24–26], and Araucariaceae [27]. Apparently, 13-epimanool is common to Pinaceae; manool, to Cupressaceae, Podocarpaceae, and Araucariaceae. However, both are reported to co-occur in *Picea ajanensis* [13] and in southern pine (*Pinus* spp.) tall oil [28]. Manool or 13-epimanool has been reported from Canadian tall oil [29], tobacco [30], and sage [31].



(**1**) Manool



(**2**) Epimanool

These diastereoisomeric compounds are difficult to distinguish because their physical, spectral, and chromatographic properties are similar [32]. The 3,5-dinitrobenzoate derivatives have been used to differentiate the two based on melting point and mixture melting point data. Unfortunately, many of the reports do not give sufficient data to distinguish which of the two compounds was isolated. Table 1 lists the best of various physical constants reported for manool, 13-epimanool, and their derivatives. Both manool and 13-epimanool can be converted to the same trihydrochloride, mp 125° [33].

Table 1. Physical constants of manool, 13-epimanool and derivatives

Derivative	Manool	13-Epimanool
Free alcohol		
Mp	54–55° [13]	36.5–38.5° [32]
$[\alpha]_D$	+33.3° [13]	+51° [32]
3,5-Dinitrobenzoate		
Mp	103–104.5° [21, 28]	119–120° [14]
$[\alpha]_D$	+8.8° [21]	+36.6° [14]
Acetate		
$[\alpha]_D$	+30° [37]	+33.8° [38]

Although the NMR spectra of manool and 13-epimanool are identical in $CDCl_3$ [32], the spectra obtained in CCl_4 have slight differences in the vinylidene portion of the spectra [4, 34] (Fig. 1). However, these differences are insufficient for calculating the epimeric purity of manool-13-epimanool mixtures.

Chiral shift reagents are useful in differentiating enantiomeric compounds by NMR [35, 36]. We find that the NMR chiral shift reagents *tris*[3-(trifluoromethylhydroxymethylene)-*d*-camphorato] europium (III), Eu(tfc)$_3$, and *tris*[3-(trifluoromethylhydroxymethylene)-*d*-camphorato] praseodymium (III), Pr(tfc)$_3$, are useful for readily differentiating manool and 13-epimanool for they allow epimeric purity to be calculated directly.

RESULTS AND DISCUSSION

As expected, Eu(tfc)$_3$ caused a downfield shift in resonance frequencies of protons spacially near the C-13 hydroxyl group of manool and 13-epimanool; Pr(tfc)$_3$, an upfield shift in these resonance frequencies. The shifts in resonance frequencies most readily observed are those for the C-13 methyl protons, the vinylidene protons, and the vinyl protons.

The shifts in resonance frequencies observed on adding Eu(tfc)$_3$ to CCl_4 solutions of manool and 13-epimanool are easily interpreted because the NMR spectra are

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* Maintained in cooperation with the University of Wisconsin.

† Systematic nomenclature follows *The Common and Systematic Nomenclature of Cyclic Diterpenes* (with Addenda and Corrigenda, Feb. 1969), proposals of a committee chaired by Dr. J. W. Rowe, Forest Products Laboratory, USDA, FS, Madison, WI 53705, U.S.A., 1968.

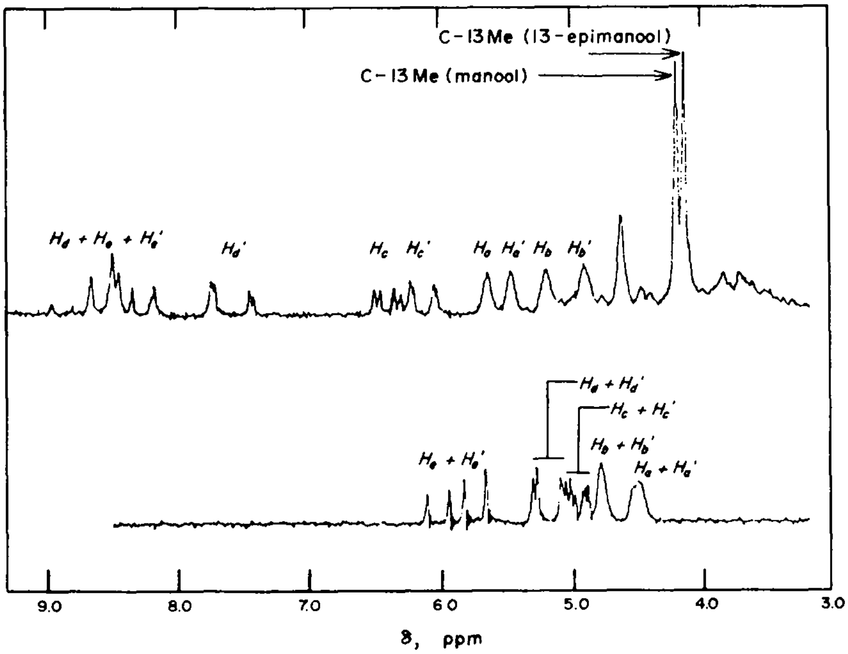

Fig. 1. Lowfield portion of NMR spectra: lower spectrum, mixture of manool (16.9 mg) and 13-epimanool (16.8 mg); and upper spectrum, the mixture with $Eu(tfc)_3$ (54 mg) added. Chemical shift of the C-13 methyls before addition of $Eu(tfc)_3$ was δ 1.20. For assignment of protons, see **1** and **2**.

spread out downfield (Fig. 1). The changes in chemical shifts for the vinylidene, vinyl, and methyl protons of manool and 13-epimanool are shown in Figs. 2 and 3.

The shifts in resonance frequencies from adding $Pr(tfc)_3$ were as large as those from adding $Eu(tfc)_3$ (Table 2). However, changes in the chemical shifts for all the various protons could not be determined with each addition of $Pr(tfc)_3$; as the vinyl and the vinylidene resonance frequencies shifted upfield they overlapped and eventually were buried in the methylene region (*ca* δ 1.3–2.7). Thus, $Eu(tfc)_3$ was preferable for differentiating manool and 13-epimanool.

These methods afford a rapid means not only of differentiating between manool and 13-epimanool but also of estimating the epimeric composition of isolated mixtures without the formation of various chemical derivatives.

Pure 13-epimanool was first isolated from *Pinus contorta* bark [32]. However, this was obtained by crystallization in low yield from a chromatographic fraction. The oily mother liquor from this fraction has been examined by these methods, and contains only 13-epimanool. The oily compound isolated from *Tsuga heterophylla* wood [19] was also shown to be epimerically pure 13-epimanool by these methods. The material isolated from southern pine (*Pinus* spp.) tall oil contained 72% manool and 28% 13-epimanool; this agrees closely with the composition previously reported from optical rotation values [28].

Table 2. Changes in chemical shifts (Δδ) on addition of $Pr(tfc)_3$ to CCl_4 solutions of manool and 13-epimanool

Molar ratio				−Δδ (ppm)*				
$Pr(tfc)_3$: (epi)manool	C-4 Me_2	C-10 Me	C-13 Me	H_a,H_a'	H_b,H_b'	H_c,H_c'	H_d,H_d'	H_e,H_e'
			Manool			*ca*	*ca*	*ca*
0.09	0.02 & 0.07	0.31	1.91	0.78	0.30	0.55	1.80	1.22
0.21	0.04 & 0.13	0.50	3.14	1.28	0.49	1.17	2.91	2.84
0.39	0.03 & 0.13	0.67	4.47	1.79	0.70	1.66		
0.78	0.04 & 0.16	0.90	6.00	2.36	0.91	2.19		
1.22	0.04 & 0.17	1.00	6.90		1.21	2.53		
			13-Epimanool					
0.09	0.04 & 0.06	0.23	1.62	0.66	0.13	0.46	1.52	1.53
0.19	0.09 & 0.11	0.38	2.76	1.07	0.25	0.98	2.40	2.48
0.38	0.11 & 0.14	0.54	3.98	1.56	0.33	1.44		
0.79	0.14 & 0.19	0.68	5.42	2.08	0.44	2.01		
1.17	0.15 & 0.21	0.78	6.18	2.35	0.49	2.28		

* For assignment of protons, see **1** and **2**. For chemical shifts and line shapes of vinyl and vinylidene protons, see Fig. 1. Chemical shifts for methyl proton singlets before addition of $Pr(tfc)_3$ are manool: C-4 Me_2 δ 0.82 and 0.87, C-10 Me 0.67, C-13 Me 1.20, and 13-epimanool: C-4 Me_2 δ 0.80 and 0.87, C-10 Me 0.67, C-13 Me 1.20.

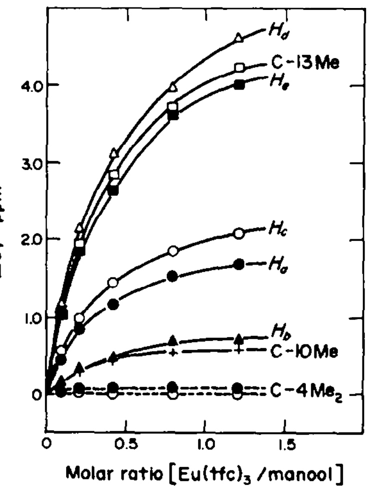

Fig. 2. Change in chemical shift ($\Delta\delta$) for various protons of manool after adding Eu(tfc)$_3$. For proton assignments see **1**.

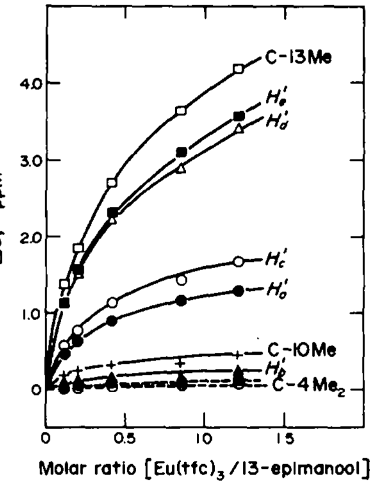

Fig. 3. Change in chemical shift ($\Delta\delta$) of various protons of 13-epimanool on adding Eu(tfc)$_3$. For proton assignments, see **2**.

EXPERIMENTAL

NMR spectra were obtained at 60 Mhz with CCl_4 solns of authentic (+)-manool and (+)-13-epimanool using TMS as the internal standard. Commercial preparations of Eu(tfc)$_3$ and Pr(tfc)$_3$ were used without further purification.

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