Interactions of Coumarins with a Lanthanide Shift Reagent: Determination of Substitution Pattern

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The effects of $Eu(fod)_3$ on the ^1H n.m.r. spectra of 22 coumarins with alkyl, alkoxy, furano, and pyrano substituents and of coumarin itself have been studied, and a method for obtaining information on the substitution pattern of the coumarins has been developed.

THE frequent occurrence and structural diversity of natural coumarins [1] has led to the application of a variety of n.m.r. techniques to their structure elucidation, particularly the use of chemical shifts and coupling,[2] the internal nuclear Overhauser effect,[3,4] and recently ^{13}C n.m.r.[5-7] Some of these techniques are not readily available, and even when they are, the unambiguous structure determination of certain types of coumarin still presents problems.

We have described briefly [8] a simple, rapid, non-degradative, non-empirical method for the structure elucidation of coumarins using the n.m.r. shift reagent $Eu(fod)_3$. We explain here how it can be used as a practical method, when the method is most useful, and also where difficulties may be encountered.

To evaluate the approach fully we have used a variety of natural and synthetic coumarins (1)—(23) with different functional groups and substitution patterns.

METHOD

A full discussion of the use of lanthanide shift reagents (l.s.r.) in n.m.r. spectroscopy has been published [9] and an earlier review may also be found helpful.[10]

It is essential that the solvent should not complex with the shift reagent, which reduces the choice, but we have found $CDCl_3$ ideal for coumarins when using $Eu(fod)_3$, and have had no reason to consider alternatives. For use in a standard 60 MHz spectrometer, we have dissolved coumarin (*ca.* 60 mg) in $CDCl_3$ (0.3 ml) containing tetramethylsilane as internal standard. The normal spectrum was obtained, and then $Eu(fod)_3$ (*ca.* 10 mg) added, dissolved, and the spectrum re-run. In each case the shifts of all the protons were measured relative to tetramethylsilane, so that the lanthanide-induced shift (l.i.s.) could be obtained. Thus, $\delta H_{[Eu(fod)_3]} - \delta H_{(untreated)} = l.i.s.$ In our approach the l.i.s. for each proton was then divided by the l.i.s. for the proton attached to C-3 of the coumarin. This gave a series of shift ratios which are independent of the weight of coumarin or shift reagent, and can be used to determine the substitution pattern of the coumarin.

We chose to use several additions (usually four) of shift reagent, and to plot the induced shift against the integral of the main resonance of the shift reagent, so as to obtain a series of straight lines which can be used to determine the shift ratios. This approach has advantages where there is overlapping of peaks, either in the original spectrum or after addition of $Eu(fod)_3$, when the precise position of a peak may be difficult to determine. In simple cases there is no advantage in the graphical approach, although more than one addition of $Eu(fod)_3$ is advisable, so that average shift values may be obtained as a precaution against random error.

Coumarins normally give very sharp resonances with good resolution, but addition of large amounts of shift reagent almost always broadens the lines. Thus there may be an advantage in using smaller amounts of coumarin initially, where instrument performance permits, so that less l.s.r. is needed. Concentrated solutions of the coumarin should be avoided for this reason.

Having obtained a series of shift ratios by either approach, there are two ways in which the data can be used. As a quick method, we have calculated (Tables 2—4) the shift ratios expected using average † complexation parameters for H, CH_3, pyran, and furan substituents in all positions (except, of course, C-3). Thus a comparison of the experimental ratios with those in Tables 2—4 may often give valuable structural information, sometimes amounting to total structure elucidation.

If it is considered desirable to refine the calculated figures for the specific coumarin under study, as we have done,[4,11,12] it is necessary to adjust ϕ and d (Figure 1) for an optimum fit to the experimental data.

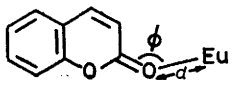
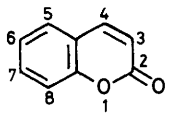


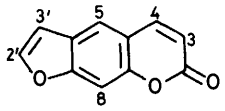
FIGURE 1

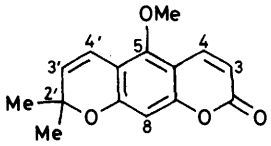
In our experience the easiest way to do this is to use a field map [13] and a Dreiding stereomodel. With the model flat on the map the carbonyl oxygen is placed on the mid-line of the field map. Movement along this line varies d, and rotation of the model in the plane of the map with the carbonyl oxygen always on the mid-line, varies ϕ. Rapid

† Three coumarins (21)—(23) were excluded from the averaging (see Discussion section).

[1] B. E. Nielsen, *Dansk. Tidsskr. Farm.*, 1970, **44**, 111; R. D. H. Murray, *Aromatic Heteroaromatic Chem.*, 1976, **4**, 422.
[2] W. Steck and M. Mazurek, *Lloydia*, 1972, **35**, 418.
[3] T. Tomimatsu, M. Hashimoto, T. Shingu, and K. Tori, *Chem. Comm.*, 1969, 168; *Tetrahedron*, 1972, **28**, 2003.
[4] A. I. Gray, R. D. Waigh, and P. G. Waterman, *J.C.S. Perkin I*, 1975, 488.
[5] Ching-jer Chang, H. G. Floss, and W. Steck, *J. Org. Chem.*, 1977, **42**, 1337, and references cited.
[6] K. K. Chan, D. D. Giannini, A. H. Cain, J. D. Roberts, W. Porter, and W. F. Trager, *Tetrahedron*, 1977, **33**, 899.
[7] D. Bergenthal, K. Szendrei, and J. Reisch, *Arch. Pharm.* 1977, **310**, 390.
[8] A. I. Gray, R. D. Waigh, and P. G. Waterman, *J.C.S. Chem. Comm.*, 1974, 632.
[9] A. F. Cockerill, G. L. O. Davies, R. C. Harden, and D. M. Rackham, *Chem. Rev.*, 1973, **73**, 553.
[10] P. V. DeMarco, *Lloydia*, 1972, **35**, 362.
[11] F. Fish, A. I. Gray, R. D. Waigh, and P. G. Waterman, *Phytochemistry*, 1976, **15**, 313.
[12] A. I. Gray, R. D. Waigh, and P. G. Waterman, *Phytochemistry*, 1977, **16**, 1017.
[13] R. M. Wing, T. A. Early, and J. J. Uebel, *Tetrahedron Letters*, 1972, 4153.



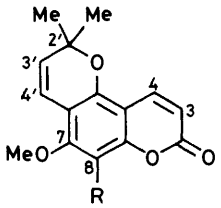




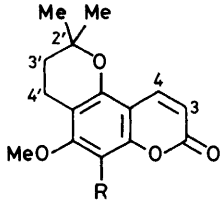
Substituent						
	3	4	5	6	7	8
(1)	H	H	H	H	H	H
(2)	H	H	H	H	OMe	H
(3)	H	H	H	Me	H	H
(4)	H	Me	H	H	OMe	H
(5)	H	Me	H	OMe	H	H
(6)	H	H	OMe	H	OMe	H
(7)	H	H	H	OMe	OMe	H
(8)	H	Me	H	H	OMe	Me

Substituent		
	5	8
(9)	H	OMe
(10)	OMe	H
(11)	H	$OCH_2CH{=}C(Me)_2$

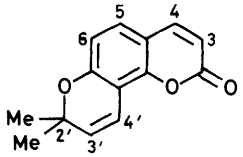
(12)



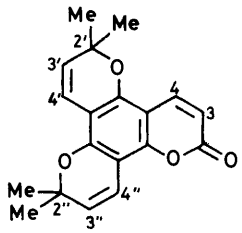
(13) R = *trans*-CH=CH·C(OSiMe$_3$)Me$_2$
(14) R = *cis*-CH=CH·C(OSiMe$_3$)Me$_2$
(15) R = *trans*-CH=CH·C(OH)Me$_2$
(16) R = *cis*-CH=CH·C(OH)Me$_2$
(17) R = *trans*-CH=CH·C(Me)=CH$_2$
(20) R = $CH_2 \cdot CH{=}CMe_2$



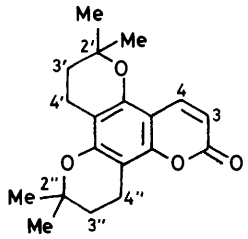
(18) R = $CH_2CH_2C(H)Me_2$
(19) R = $CH_2CH_2C(OH)Me_2$



(21)



(22)



(23)

visual inspection of proton positions on the map will then reveal if the shift ratios are reasonable for the compound in question. When a ' best fit ' has been obtained, the proton positions can be marked on the field map so that θ and R can be measured for each one (Figure 2).

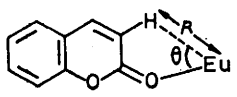


FIGURE 2

The value of $(3\cos^2\theta - 1)/R^3$ can then be calculated for each proton to give the relative shifts. For comparison with the experimental figures, the relative shifts must be normalised to relative shift 3-H ≡ 1.00, and this is done by dividing each relative shift figure by the value of $(3\cos^2\theta - 1)/R^3$ obtained for 3-H. To save time on the calculations we plotted $3\cos^2\theta - 1$ for all possible values of θ, so that values did not need to be calculated individually.

Where substituents are not in the plane of the ring, as with methylpyran and methoxy (see Discussion section) measurements of θ and R are more awkward, but once ϕ and d have been chosen with substituents which do fall in the plane of the field map the problem is simplified.

The experimental and calculated values obtained as above for all the coumarins are given in Table 1.*

* Non-systematic numbering has been used wherever necessary to facilitate comparisons between simple and polycyclic coumarins.

DISCUSSION

Normally correlations of structure with l.i.s. are three-dimensional problems, for which a computer is required to calculate the direction and length of the lanthanide–ligand bond which give a ' best fit ' to the observed data. The advantage of the present method is that the problem of location of the europium atom has been reduced to two dimensions so that a computer is not necessary. While this involves assumptions about the bonding of coumarins to $Eu(fod)_3$ which may not be universally valid, a good fit has been obtained for a large number of coumarins for all substituents (Table 1), and the method has been applied successfully.[4,11,12,14] Theoretical calculations of the electron densities in coumarins also tend to support our findings.[15]

The only examples in which a poorer than usual fit was obtained were those with 7,8-pyran substitution [seselin (21), dipetalolactone (22), and tetrahydrodipetalolactone (23)]. For these three compounds the 4-H shift was less than calculated if the ϕ and d values were altered to give a good fit elsewhere, and the ϕ and d values thus obtained were atypical (Table 1). Possibly a better fit could be obtained by allowing movement of the europium atom or the ring substituents out of the plane of the coumarin ring or to allow the ring to become non-planar, but the present method could not then be used.

Some coumarins [*e.g.* (7), (15), (16), (19)] possess additional complexation sites for $Eu(fod)_3$. Obviously the presence of such a site destroys the simple relationship between induced shift and substituent position which is the basis of the method. Where the additional site is a side-chain hydroxy, as in *cis*- and *trans*-avicennol,[4,12] it can be conveniently protected as the trimethylsilyl ether. In other cases the method of protection would have to be devised specifically, but a special problem may be posed by the presence of *o*-methoxys. An isolated methoxy does not complex significantly with $Eu(fod)_3$, but *o*-methoxys do,[9] and the difference has been used to confirm the structure of 2,4,5-trimethoxystyrene.[16] Similar complexation occurs with the methoxys of scoparon (7) and it is clear from the figures in Table 1 that the relative l.i.s. for all protons in scoparon (except by definition 3-H) are much larger than usual. Subtraction of average values for shifts in each position from Table 2, calculated for complexation at the lactone carbonyl only, gives an

TABLE 1

Observed and calculated shift values for coumarins, relative to 3-H ≡ 1.00 (calculated values in parentheses)

			Substituent position							
Compound	ϕ(°)	d/Å	4	5	6	7	8	2′	3′	4′
Coumarin (1)	156	3.00	0.31(0.32)	0.19(0.19) [a]	0.15(0.14) [a]	0.19(0.17) [a]	0.31(0.33)			
Herniarin (2)	159	3.30	0.34(0.34)	0.20(0.20)	0.16(0.15)	0.10(0.12)	0.29(0.32)			
6-Methylcoumarin (3)	158	3.25	0.29(0.32)	0.20(0.18)	0.10(0.11)	0.15(0.16)	0.27(0.31)			
4-Methylherniarin (4)	160	2.60	0.22(0.20)	0.22(0.20)	0.18(0.15)	0.12(0.12)	0.34(0.34)			
6-Methoxy-4-methylcoumarin (5)	160	2.60	0.20(0.20)	0.20(0.20)	0.11(0.11)	0.18(0.17)	0.33(0.34)			
Limettin (6)	158	3.25	0.31(0.32)	0.09(0.10) [c]	0.16(0.14)	0.13(0.14) [c]	0.28(0.31)			
Scoparon (7)	*b*		0.40	0.54	0.36	0.36	0.61			
4,8-Dimethylherniarin (8)	160	2.63	0.22(0.20)	0.22(0.20)	0.18(0.15)	0.13(0.12)	0.30(0.29)			
Xanthotoxin (9)	145	2.75	0.25(0.26)	0.20(0.18)			0.29(0.29)	0.10(0.10)	0.11(0.10)	
Bergapten (10)	158	3.45	0.34(0.33)	0.14(0.13)			0.30(0.32)	0.11(0.09)	0.12(0.11)	
Imperatorin (11)	142.5	2.50	0.31(0.30)	0.22(0.22)			*d*	0.12(0.12)	0.14(0.13)	
Xanthoxyletin (12)	159.5	3.35	0.30(0.33)	0.17(0.14)			0.30(0.32)	0.07(0.09)	0.05(0.08)	0.11(0.12)
trans-Avicennoltrimethylsilyl ether (13)	152.5	2.75	0.31(0.31)			0.15(0.15)	*d*	0.08(0.08)	0.08(0.08)	0.13(0.12)
cis-Avicennoltrimethylsilyl ether (14)	152.5	2.75	0.32(0.31)			0.16(0.15)	*d*	0.08(0.08)	0.09(0.08)	0.15(0.12)
trans-Avicennol (15)	*b*		0.40			0.40	*d*	0.14	0.14	0.26
cis-Avicennol (16)	*b*		0.35			0.29	*d*	0.09	0.08	0.18
Avicennin (17)	151	2.80	0.29(0.30)			0.13(0.13)	*d*	0.08(0.08)	0.08(0.08)	0.12(0.11)
Hexahydroavicennin (18)	151	2.80	0.30(0.30)			0.13(0.13)	*d*	0.09(0.08)	0.11(0.09)	0.13(0.12)
Tetrahydroavicennol (19)	*b*		0.30			0.60	*d*	0.12	0.20	0.41
Dipetaline (20)	152.5	2.75	0.31(0.31)			0.15(0.15)	*d*	0.08(0.08)	0.08(0.08)	0.12(0.11)
Seselin (21)	184	2.40	0.30(0.37)	0.20(0.21)	0.13(0.14)			0.05(0.06)	0.04(0.05)	0.23(0.23)
			4	2′	3′	4′	2″	3″	4″	
Dipetalolactone (22)	184	2.40	0.31(0.37)	0.08(0.10)	0.07(0.08)	0.09(0.10)	0.05(0.06)	0.03(0.05)	0.22(0.23)	
Tetrahydrodipetalolactone (23)	184	2.40	0.29(0.37)	0.07(0.11)	0.07(0.11)	0.08(0.11)	0.04(0.07)	0.05(0.07)	0.19(0.24)	

[a] Assignments uncertain owing to complex splitting and overlapping. [b] Two complexation sites. [c] See text. [d] See Table 5.

TABLE 2

Calculated shifts for simple substituents in all positions (θ 156°, d 3.0 Å), relative to 3-H ≡ 1.00

	Position				
Substituent	4	5	6	7	8
H	0.32	0.19	0.14	0.17	0.33
Me	0.19	0.15	0.11	0.13	0.29
OMe(C)	0.13	0.17	0.10	0.11	0.16
OMe(A)	0.16	0.11	0.10	0.15	0.50
OMe(O)	0.15	0.13	0.10	0.12	0.26

C = Clockwise, A = anticlockwise, O = out-of-plane (coumarin as drawn in this paper).

[14] K. K. Purushothaman, S. Vasanth, J. D. Connolly, and C. Labbé, *J.C.S. Perkin I*, 1976, 2594.

[15] C. Decoret and J. Royer, *Bull. Soc. chim. France*, 1976, 587; M. Abou-Asrali, C. Decoret, J. Royer, and J. Dreux, *Tetrahedron*, 1976, **32**, 1655.

[16] P. G. Waterman, *Phytochemistry*, 1976, **15**, 347.

TABLE 3

Calculated shifts for dimethylpyranocoumarins (ϕ 156°, d 3.0 Å), relative to 3-H ≡ 1.00

Ring fusion	Substituent 2′,2′-Me_2	3′-H	4′-H
6,5	0.08	0.08	0.16
5,6	0.09	0.08	0.11
7,6	0.09	0.08	0.11
6,7	0.09	0.09	0.14
8,7	0.16	0.11	0.12
7,8	0.10	0.13	0.33

TABLE 4

Calculated shifts for furanocoumarins (ϕ 156°, d 3.0 Å), relative to 3-H ≡ 1.00

Ring fusion	Substituent 2′-H	3′-H
6,5	0.09	0.15
5,6	0.09	0.11
7,6	0.09	0.10
6,7	0.09	0.13
8,7	0.13	0.12
7,8	0.13	0.31

approximate measure of the effect of *o*-methoxy complexation with scoparon (Figure 3). In a recent con-

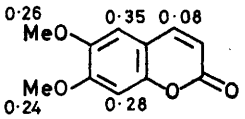


FIGURE 3

trasting example only simple complexation with a coumarin occurred,[14] despite the presence of 6,7-dimethoxy substitution. Clearly some further work needs to be done before the effect of molecular structure on

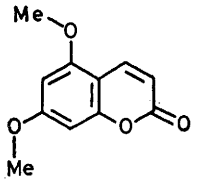
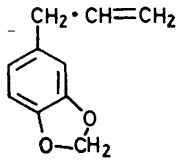


FIGURE 4

methoxy complexation is fully understood. Complexation with *m*-methoxys does not appear to be significant, as shown by the results for limettin (6) (Table 1). No



(24)

methylenedioxy-substituted coumarins were readily available to us, but experiments with safrole (24) indicate that complexation between $Eu(fod)_3$ and the methylenedioxy group would be weak and unlikely to interfere with the method.

In most of our work we have assumed that methoxy substituents would adopt an out-of-plane conformation in order to minimise steric interactions with groups *ortho* to them. Where the *ortho*-substituents are small this may not be valid, results in other areas [17] suggesting that there is a favourable conformation for the methyl in the plane of the ring, stabilised by overlap of the lone pairs of the ether oxygen with the aromatic π-electrons. The good fit we have obtained in most cases suggests that the out-of-plane conformation is a reasonable approximation, except where a substituent on one side of the methoxy dictates a move towards the less hindered side. Thus in the case of limettin (6) the observed shift for what we assume to be the 5-methoxy is lower than calculated, unless rotation towards C-6 away from 4-H is invoked (Table 2). This may cause rotation of the 7-methoxy away from the 5-methoxy, resulting in a conformation which fits the observed shifts (Figure 4). Clearly further work would be needed to justify these refinements, but there is a probability that low values may be observed for 5-methoxys in coumarins lacking 6-substitution, for this reason.

TABLE 5

Shifts for 8-isoprenyl substituents (from point of attachment to coumarin) [a]

Compound	Chemical shift (3-H ≡ 1.00)			
(11)	CH_2 0.29	CH 0.21	Me_2 0.13, 0.07	
(13)	CH 0.40(0.33)	CH 0.52(0.45)	$SiMe_3$ 0.11(0.11)	Me_2 0.13(0.12)
(14)	CH 0.32	CH 0.25	$SiMe_3$ 0.19	Me_2 0.28
(15)	CH 2.08	CH 2.56	OH 8.57	Me_2 1.59
(16)	CH 0.66	CH 0.74	OH 3.78	Me_2 0.81
(17)	CH 0.41(0.32)	CH 0.38(0.45)	Me 0.09(0.15)	$=CH_2$ 0.09, 0.03(0.15, 0.08)
(18)	CH_2 0.36	CH_2 0.24	CH [b]	Me_2 0.11
(19)	CH_2 2.92	CH_2 4.03	OH 7.35	Me_2 2.17
(20)	CH_2 0.35(0.32)	CH 0.54(0.48)	Me_2 0.08, 0.19(0.12, 0.15)	

[a] Calculated values, where given in parentheses, are for simple averages of in-plane conformations. [b] Not identified.

While the calculated figures in Table 2 refer to a limited number of substituents, it is clear that the figures for CH_3 and OCH_3 will serve as a reasonable approximation for hydrogens attached to the first carbon of longer chains. The flexibility of longer chains is such as only to allow estimates for the predicted shifts, but particularly where an 8-substituent is concerned shift differences are large enough to allow the positional assignment to be made with fair confidence.[4,11,12] In Table 5 we have given the observed and, in three cases, calculated values for flexible prenyl-derived side-chains in all the compounds we have had

[17] A. Hofer, *Tetrahedron Letters*, 1975, 3415.

the opportunity to evaluate. Where given, the calculated values are based on average values for two extreme conformations, so that only an approximate fit would be expected. In some cases extra information can be obtained from simplification of splitting patterns in the side chain, and even configurational information may be obtained.[12]

The relatively poor fit obtained for 7,8-pyranocoumarins may be tentatively explained by means of steric interactions between a relatively inflexible 8-substituent and the lactone ring oxygen. For such compounds the method still offers useful structural information on a more empirical basis, since the shift for the 8-substituent is larger than would be obtained elsewhere. Further work is in progress in an attempt to clarify this anomaly.

EXPERIMENTAL

Spectra were recorded on a Perkin-Elmer R12 n.m.r. spectrometer. Coumarins were obtained from a variety of natural and commercial sources, or by routine methylation of commercially available hydroxycoumarins. Structures were confirmed by comparison with literature data (u.v., i.r., n.m.r., m.s., m.p.) except for new compounds which were identified previously.[4,11,12]

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