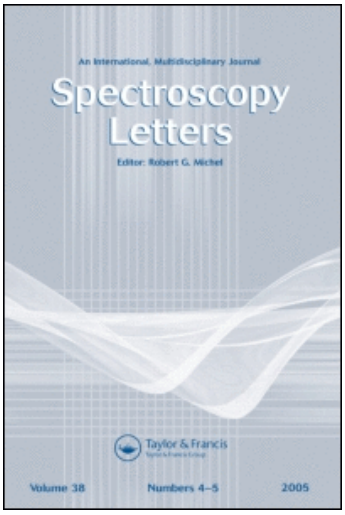

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^{1}H NMR Studies of Drugs with Achiral and Chiral Lanthanide Shift Reagents. CGP29953: Analog of the Aromatase Inhibitor, Aminoglutethimide

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^{1}H NMR STUDIES OF DRUGS WITH ACHIRAL AND CHIRAL LANTHANIDE SHIFT REAGENTS. CGP29953: ANALOG OF THE AROMATASE INHIBITOR, AMINOGLUTETHIMIDE.

Key Words: Anti-cancer agent; Stereoisomers; Enantiomers; Eu(FOD)$_3$; Eu(HFC)$_3$; 1-(4-Aminophenyl)-3-n-propyl-3-azabicyclo[3.1.0]hexane-2,4-dione; One- and two-dimensional ^{1}H NMR.

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ABSTRACT

1-(4-Aminophenyl)-3-n-propyl-3-azabicyclo[3.1.0]hexane-2,4-dione, compound 1 (CGP29953), is an analog of the aromatase inhibitor aminoglutethimide (AG); AG has applications in cancer treatment. We have studied the ^{1}H NMR spectra of racemic compound 1 (in $CDCl_3$, ambient temperatures, 300 MHz) with the added achiral LSR, tris(6,6,7,7,8,8,8-heptafluoro-2,2-dimethyl-3,5-octanedionato)europium(III), compound 2, and the chiral LSR, tris[3-(heptafluoropropylhydroxymethylene)-(+)-camphorato]europium (III), compound 3. Use of compound 3

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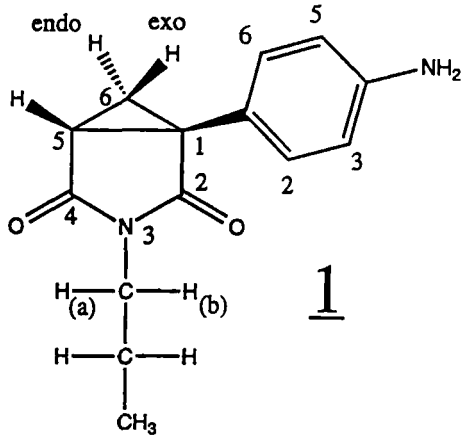
produced substantial enantiomeric shift differences for several nuclei of compound 1. ^{1}H-^{1}H COSY spectra were used to determine the relative sense of magnetic nonequivalence for selected nuclei of compound 1 with added compound 3. Relative lanthanide induced shift (LIS) magnitudes for compound 1 with added LSR are discussed.

INTRODUCTION

Aminoglutethimide, AG, 3-(4-aminophenyl)-3-ethylpiperidine-2,6-dione, has played a significant role in some treatments for hormone-dependent cancers because of its activity as an inhibitor of the enzyme, aromatase. One of the enantiomers of AG has substantially greater potency [1,2]. Many analogs of AG have been prepared and examined for potential greater potency and selectivity.

For some time, our lab has investigated the use of achiral and chiral lanthanide shift reagents (LSR) for NMR studies of drugs and analogs. General methods and principles of LSR have been discussed [3]. We have published NMR studies based on LSR techniques for glutethimide [4] and aminoglutethimide [5]. In addition, we have reported work on the chiral piperidinedione, thalidomide [6], and on a series of chiral substituted pyrrolidinediones, including ethosuximide [7], methsuximide [8], phensuximide [9], α-ethyl-α-phenylsuccinimide [10], and fenimide [11].

The compound 1, CGP29953, 1-(4-aminophenyl)-3-n-propyl-3-azabicyclo[3.1.0]hexane-2,4-dione, had been synthesized and evaluated for activity toward the aromatase and cholesterol side chain cleavage (CSCC) enzymes [12]. Subsequently, the compound was reported as a substrate in studies with chiral stationary phases for liquid chromatography [13]. Recently, some methods of capillary electrophoresis have been applied to the chiral resolution of compound 1 [14]. Compound 1 is structurally a close analog of AG. Some related "cyclopropanedicarboximides", i.e., possessing the 1-aryl-3-azabicyclo[3.1.0]hexane-2,4-dione system, had also been prepared and studied [15]. NMR methods for enantiomeric excess (% e.e.) determination employing β-cyclodextrin as a



Structure of compound 1.

chiral solvating agent had been applied to another AG analog [16]. We were interested in exploring the use of LSR using a medium field NMR spectrometer with compound 1 for several reasons. Compared to AG, construction of the cyclopropane ring to form the bicyclo [3.1.0] ring system in compound 1 produces a much more rigid molecule. In addition, compound 1 formally may be regarded as a pyrrolidinedione (succinimide) ring system related to several we have examined earlier [7-11]. As with other analogs possessing multiple potential LSR binding sites, we wanted to qualitatively evaluate preferred binding sites in compound 1. Increased rigidity in compound 1 (relative to AG) as a substrate for LSR might be expected to reduce the number of possible contributing bound complexes with differing conformers, and thus might enhance observed enantiomeric shift differences ($\Delta\Delta\delta$). The potential intrinsic importance of compound 1, as well as the earlier studies [12-14] also increased the relevance of these investigations. We planned to examine compound 1 with the achiral LSR, tris(6,6,7,7,8,8,8-heptafluoro-2,2-dimethyl-3,5-octanedionato)europium(III), compound 2, and with the chiral

LSR, tris[3-(heptafluoropropylhydroxymethylene)-(+)-camphorato]europium(III), compound 3.

EXPERIMENTAL

Racemic compound 1 (CGP29953) was kindly provided by Ciba-Geigy (reported mp 111-112°) and was used without further purification. General methods followed those reported earlier [6,8,9]. Spectra were obtained in $CDCl_3$ solution [containing 0.03% $(CH_3)_4Si$ as internal standard at 0.00 ppm] at ambient temperatures. A Bruker ACF300 spectrometer with QNP probe and ASPECT 3000 data system was used, for a ^1H observe frequency ca. 300 MHz. For typical 1D spectra, digital resolution of ca. 0.4-0.5 Hz/point was employed. No zero-filling or apodization was used. Standard Bruker microprograms were used for the COSY spectrum. For runs with added LSR, weighed portions of a stock solution of the LSR in $CDCl_3$ were added to the solution of compound 1. For example, a $Eu(FOD)_3$ stock solution contained 54.1 mg of compound 2 in 502.4 mg solvent $CDCl_3$. The $Eu(HFC)_3$ stock solution contained 68.9 mg of 3 in 925.8 mg $CDCl_3$ solvent. Initial concentrations of compound 1 for runs with compounds 2 or 3 were 0.040 molal. For the highest [3]:[1] ratio employed (ca. 1.79), solid LSR was added to the sample solution. When nuclei exhibited enantiomeric shift differences in the presence of chiral LSR compound 3, reported chemical shifts are the average values for the two enantiomers. Solvent and LSRs were obtained from Aldrich Chemical Co. (Milwaukee WI) and were used as supplied.

RESULTS AND DISCUSSION

The unshifted ^1H NMR reference spectra of 0.040 molal compound 1 showed the expected symmetrical pattern for the AA'BB' aminophenyl system, with approximate doublet of triplet (dt) 2H multiplets at 7.166 ppm for aryl H-2,6 and 6.668 ppm for H-3,5, reflecting shielding due to mesomeric electron release by the NH_2 group of H-3,5 [17a] based on substituent induced shifts. Observed couplings of ca. 8.48 and 2.29 Hz were seen. Confirmation of the assignments was provided with added LSR as seen by greater lanthanide-induced

shift (LIS) magnitudes and greater broadening for H-3,5. The broad 2H singlet for the NH_2 appeared at 3.737 ppm. The diastereotopic pair of NCH_2 protons, H(a) and H(b), resonated at 3.380 ppm as an approximate td pattern (with additional outer lines) with observed splittings of 7.31 and 1.77 Hz. The HCC=O resonance was assigned to the dd pattern at 2.606 ppm, ^3J = 8.09, 3.64 Hz. Deshielding by the carbonyl moves this cyclopropane hydrogen to lower field versus the ring CH_2. The larger vicinal coupling is consistent with cis couplings for cyclopropane, since dihedral angles should be near 0° [17b]. We assign the approximate dd and triplet patterns at ca. 1.780 and 1.729 ppm to the exo and endo ring methylene protons, respectively, based on apparent couplings of 8.09 and 4.47 Hz for the 1.780 ppm signal. The larger coupling, 8 Hz, is consistent with cis vicinal splitting to HCC=O of the cyclopropane, and the smaller 4.5 Hz coupling is reasonable for a ^2J(geminal) cyclopropane splitting. The near-triplet at 1.729 ppm could result from approximate isogamy, with equal couplings of ^2J(geminal) and ^3J(trans) on the cyclopropane ring; this signal is thus assignable to the endo hydrogen. Representative values of ca. +4 and -4 Hz for the ^3J trans and ^2J cyclopropane couplings respectively, appear reasonable [17b]. The observed splitting was ca. 4.07 Hz. Although the CH_2CH_3 protons are diastereotopic, they are evidently nearly isochronous in the absence of LSR, and absorb at 1.553 ppm (2H, approx. sextet) reflecting similar vicinal couplings of ca. 7.38 Hz to the neighboring NCH_2 and CH_3 protons. The highest field signal, at 0.879 ppm, is the CH_3 (3H, t, J = 7.45 Hz). The unshifted reference spectrum of compound 1 is shown in Figure 1.

Twelve increments of the achiral $Eu(FOD)_3$ were employed, up to a [2]:[1] molar ratio of 1.10. Results are shown in Figure 2. Some significant observations may be noted. The higher field aryl proton signal (in the unshifted reference spectrum of compound 1) moves downfield faster than the aryl resonance initially at lower field, leading to a "crossover" of signals as LSR is added. Greater lanthanide-induced

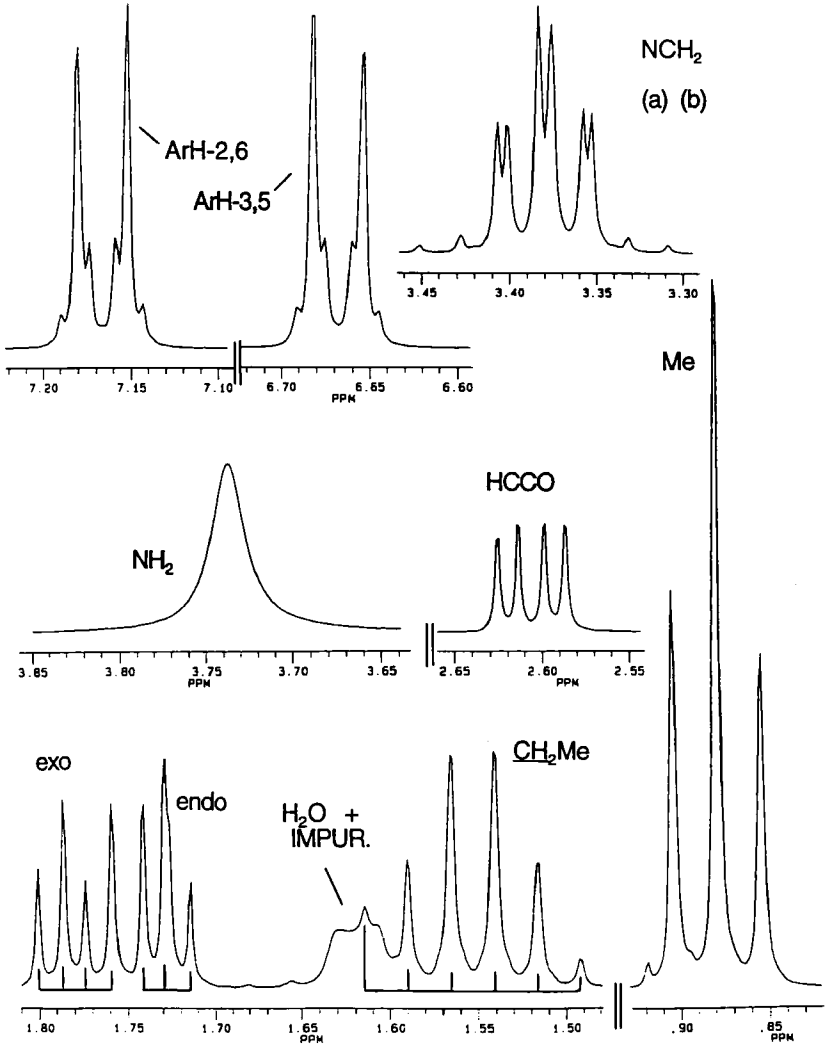


Figure 1. The unshifted reference ^1H NMR spectrum for racemic compound 1 in $CDCl_3$ (300 MHz, ambient temperature). All expansions are shown with uniform vertical (intensity) and horizontal (shift) scales. Brackets denote selected multiplets: H(exo) = double doublet; H(endo) = approx. triplet; $\underline{CH_2}Me$ = sextet.

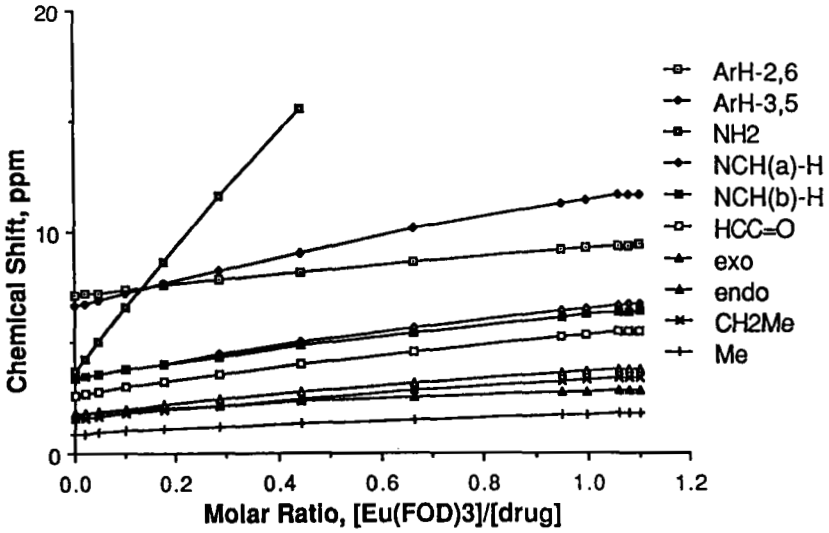


Figure 2. Variation of chemical shifts (ppm) versus [Eu(FOD)$_3$]:[1] molar ratio.

broadening is seen for the aryl signal that moves downfield faster. These results are consistent with substantial LSR binding at the NH_2, which would cause larger LIS magnitudes and line broadening for the aryl H-3,5 than for aryl H-2,6. There is also a crossover of signals for the cyclopropyl CH_2 exo/endo pair as well. The diastereotopic NCH_2 protons show clear chemical shift *nonequivalence* at high 2:1 molar ratios. Increased observed multiplicity for the CH_2CH_3 resonance as LSR is added suggests increasing anisochrony for this diastereotopic pair of methylene hydrogens, as well as for the NCH_2 and exo/endo pairs.

Spectra of compound 1 were also acquired with additions of the chiral Eu(HFC)$_3$ with LIS results shown in Figure 3. Apparent enantiomeric shift differences, $\Delta\Delta\delta$, are seen for signals of every proton except for the broad NH_2 absorption. Variations of $\Delta\Delta\delta$ with [3]:[1] molar ratios are summarized in

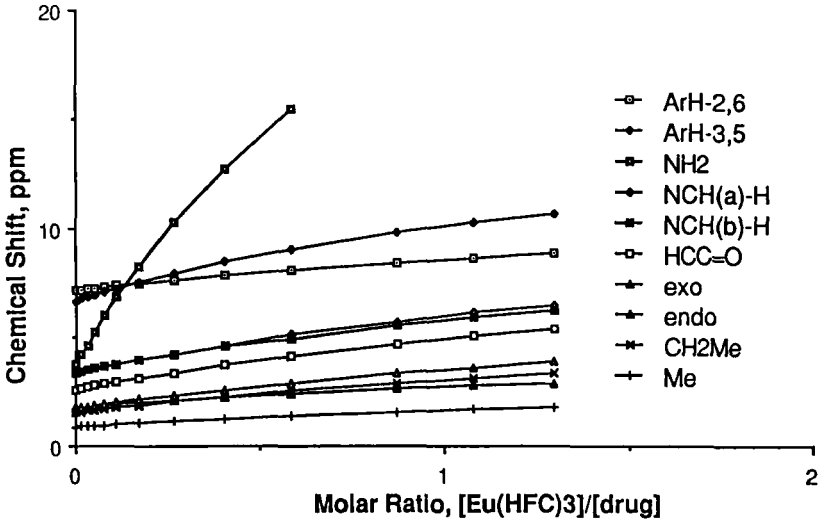


Figure 3. Variation of chemical shifts (ppm) versus [Eu(HFC)$_3$]:[1] molar ratio.

Figure 4. (The $\Delta\Delta\delta$ for the HCC=O signal is not included in Fig. 4 because, due to signal multiplicity and broadening, a clear observation and good estimate of $\Delta\Delta\delta$ could only be obtained on one 3:1 ratio [ca. 0.58].) Some potential utility is apparent for direct determination of % e.e. for samples of compound 1 with added compound 3, as seen in the selected spectral expansions of Figure 5. For several traces of Figure 5, differential lanthanide-induced line broadening is observed, e.g., for the aryl H-3,5 traces, b,c,e and f, in which the lower field enantiomer's signal appears broader. Peak area measurements (rather than peak height measurements) would be preferable for % e.e. measurements in such cases. For compound 1 with compound 3, dramatically larger $\Delta\Delta\delta$ magnitudes are seen for aryl H-3,5 than for H-2,6, despite greater proximity of the latter to the chiral center. This suggests that the greater proximity of aryl H-3,5 to a major LSR binding site at the NH_2 is of key importance. The

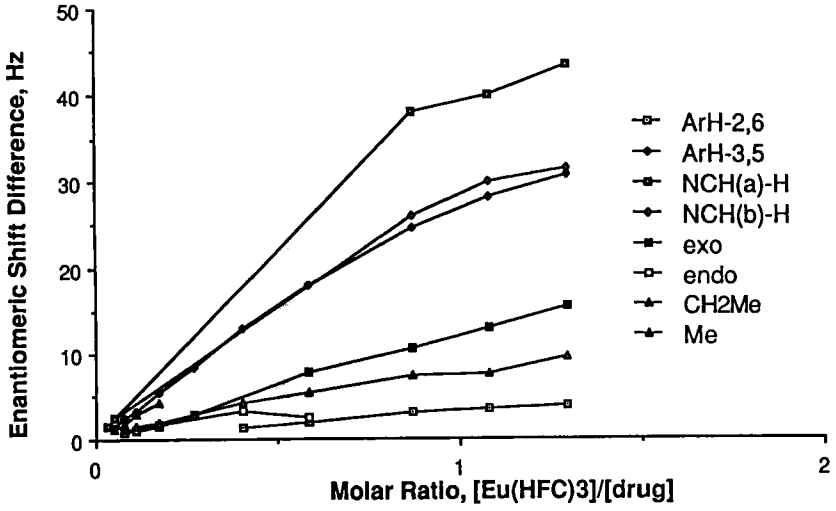


Figure 4. Variation of enantiomeric shift differences (in Hz) versus [Eu(HFC)$_3$]:[1] molar ratio.

largest $\Delta\Delta\delta$ magnitudes are seen for each of the diastereotopic hydrogens of the NCH_2, although they are not very close to the chiral center compared to, e.g., the cyclopropyl hydrogens and aryl H-2,6. We suggest that this supports appreciable contributions from bound complexes with compound 3 coordinated at the imide carbonyls, which would result in close proximity of lanthanide to the NCH_2.

Because of spectral complexity in the presence of compound 3, there were uncertainties in assignments of some signals. For example, at a 3:1 molar ratio of 1.79, with four complex multiplets distinguishable for signals of the diastereotopic NCH_2 pair, it was not immediately obvious which absorptions resulted from $\Delta\Delta\delta$ and which from diastereotopic anisochrony. The ^{1}H-^{1}H homonuclear chemical shift correlation experiment, COSY45, was employed to clarify this and to assign the relative senses of magnetic nonequivalence for some nuclei of compound 1. The resulting

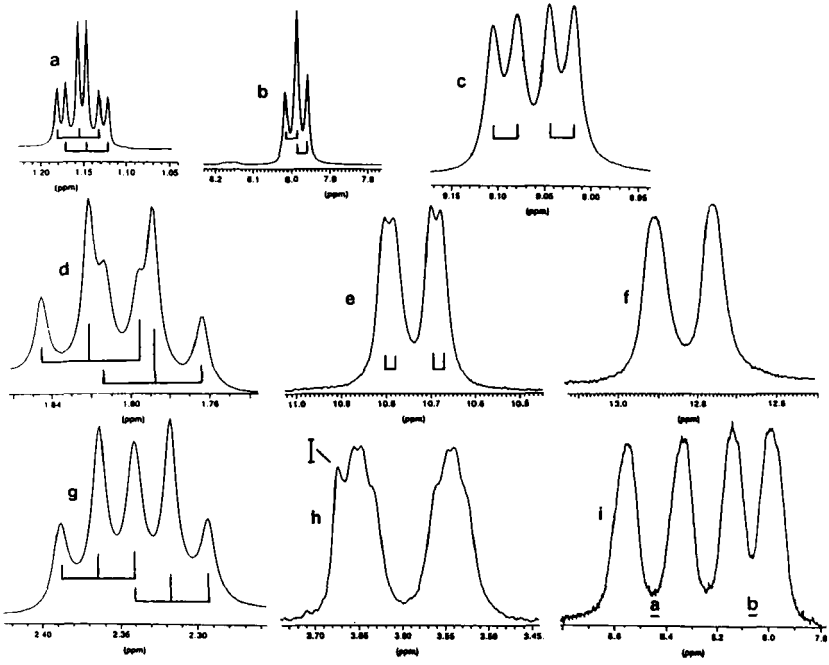


Figure 5. Selected expansions of 300 MHz ^{1}H NMR spectra of compound 1 with added LSR 3. For each trace, the assignment, [3]:[1] molar ratio, average chemical shift (ppm), and enantiomeric shift difference [Hz] are given. Note that vertical intensity and horizontal shift may differ between the traces. Where shown, brackets denote gross multiplet structure from each enantiomer.

(a) CH_3:	0.267,		1.15 ppm	[2.95 Hz],
(b) H-3,5:	0.267,		7.99 ppm	[8.46 Hz],
(c) H-3,5:	0.585,		9.06 ppm	[18.02 Hz],
(d) CH_3:	1.300,		1.80 ppm	[9.56 Hz],
(e) H-3,5:	1.300,		10.74 ppm	[30.8 Hz],
(f) H-3,5:	1.792,		12.83 ppm	[44.9 Hz],
(g) CH_3:	1.792,		2.34 ppm	[13.61 Hz],
(h) H(exo):	1.792,		3.60 ppm	[32.35 Hz] (I=impurity),
(i) NCH_2:	1.792,	H(a):	8.45 ppm	[64.7 Hz] and
		H(b):	8.07 ppm	[46.3 Hz].

spectrum is shown in Figure 6. Thus, in the region with four broad peaks ca. 7.9-8.6 ppm, crosspeak correlations are seen between alternate peaks (i.e., first to third peaks, and second to fourth peaks) rather than between adjacent peaks. The correlation implies (geminal) coupling between the diastereotopic pair NCH(a)H(b) within each single enantiomer (single molecule). Therefore, the spacing from the first to the second peak (labeling from lower to higher field) is the $\Delta\Delta\delta$ for "H(a)" since there is no crosspeak linking these signals. Similarly, the separation from the third to the fourth peak denotes $\Delta\Delta\delta$ for "H(b)". For the diastereotopic $\underline{CH_2}CH_3$, two broad overlapped multiplets are seen ca. 4.3-4.5 ppm. A priori, it is not clear whether the signal separation denotes $\Delta\Delta\delta$ or diastereotopic anisochrony. But the lower field multiplet near 4.5 ppm shows crosspeaks to only peaks 1 and 3 (at ca. 8.6 and 8.1 ppm) of the NCH_2 resonance, and the higher field multiplet at about 4.35 ppm correlates exclusively with peaks 2 and 4 of the NCH_2 signal, near 8.3 and 7.95 ppm. Since peaks 1 and 3 of the NCH_2 signal cluster are derived from a single enantiomer of compound 1, and correlate to the lower field 4.5 ppm portion of the $\underline{CH_2}CH_3$ absorptions, these are all associated with the one enantiomer. The higher field NCH_2 signals, peaks 2 and 4 (at ca. 8.3 and 8.0 ppm), both correlate to the 4.35 ppm multiplet, linking these signals as derived from the other enantiomer of compound 1. Since the lower field signals for NCH(a)H(b) correlate with the lower field $\underline{CH_2}CH_3$ absorption, all of these nuclei have the same sense of magnetic nonequivalence. The separation between the $\underline{CH_2}CH_3$ multiplets (near 4.35 and 4.5 ppm) roughly define a separation due to $\Delta\Delta\delta$; the diastereotopic anisochrony is a minor contributor. In a similar way, the COSY spectrum crosspeaks allow us to recognize the same relative sense of magnetic nonequivalence between the HCC=O signals (ca. 7.0-7.2 ppm), the endo proton signal (ca. 5.2 ppm), and the exo signal (ca. 3.6 ppm), since the lower field portions in each region are correlated with one another. This is observable even when resolution between

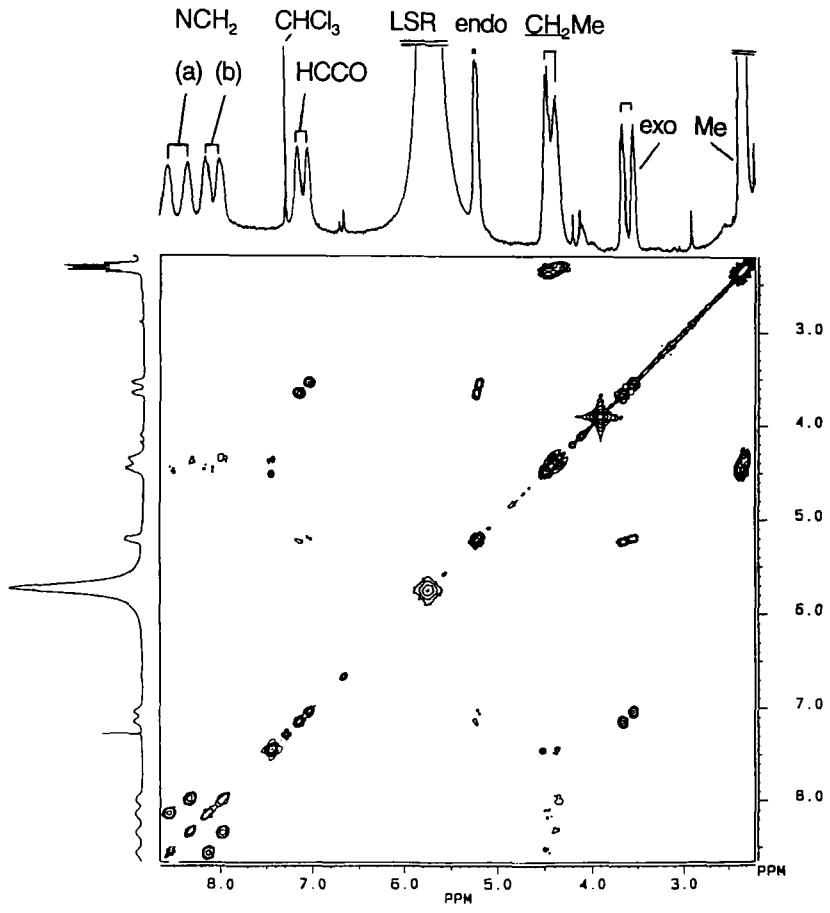


Figure 6. The ^1H-^1H homonuclear chemical shift correlation spectrum (COSY45) for compound 1 with added $Eu(HFC)_3$. The [3]:[1] molar ratio is 1.792. The spectral width in f_2 was 1938 Hz, with 256 increments in t_1, zero-filled once in f_1 and f_2 for a final data matrix of 512 x 1024. The spectrum was in the magnitude mode, with 2 dummy scans and 16 acquisitions for each t_1 increment. Data were processed with unshifted sine-bell apodization in both dimensions, and symmetrized. Some extra signals are seen on the diagonal due to aliasing. Brackets denote pairs of signals in enantiomers of compound 1.

each enantiomer's signal is limited, as for the endo absorption. Finally, the same relative sense of magnetic nonequivalence is seen for the CH_3 signals with respect to the NCH_2CH_2 absorptions. Thus, within each of the spin systems encompassed by the COSY spectrum, that is, the N-propyl system and the cyclopropyl system, all nuclei show a commonality of magnetic nonequivalence sense. Since these spin systems are not coupled to each other, however, we can not draw conclusions as to sense of magnetic nonequivalence between the two systems.

Table 1 summarizes data from Figs. 2 and 3 regarding LIS magnitudes versus [LSR]/[1] molar ratios, based on slopes from line equations for each nucleus following linear least squares line fitting, for experimental points at lower [LSR]:[drug] ratios, i.e., below 0.5. At these lower LSR levels, better linearity was observed. Correlation coefficients, R, were consistently good, with R=1.00 for all nuclei, with each LSR, over the range used. Normalized slopes were obtained from each LSR series relative to the H(endo) slope. These normalized values from runs with $Eu(FOD)_3$ and $Eu(HFC)_3$ are generally in rather good agreement, within ca. 10%. Slightly larger differences in values are seen for aryl H-2,6 [26%]; aryl H-3,5 [14%]; NH_2 [12%]; and NCH(a) [14%]. The similarity of normalized slope values, obtained with LSR 2 and LSR 3, would suggest similar contributions (of bound complexes with compounds 1 and 2, or with compounds 1 and 3) from lanthanide bound at each respective binding site. This would imply comparable LIS magnitudes for the three cyclopropyl hydrogens (part of the rigid bicyclic system) and the N-propyl group for both LSRs. We suspect that a possible significant difference in the geometries of the bound complexes of compound 1 (with LSR 2 or 3) may be the conformation of the aminophenyl ring about the aryl-C bond.

While major LSR binding appears to be at the NH_2 group, as indicated by large LIS magnitudes for the NH_2 and aryl H-3,5, substantial lanthanide binding must also be occurring at

Table 1. Slopes of calculated line equations for plots of lanthanide-induced shifts vs. molar ratios of [LSR] : [CGP29953] for nuclei of compound 1 with added LSR 2 or 3.

	$Eu(FOD)_3$ data		$Eu(HFC)_3$ data	
Nucleus	Unnorm'd	Normalized	Unnorm'd	Normalized
ArH-2,6	2.386	1.029	1.784	0.814
ArH-3,5	5.495	2.371	4.538	2.071
NH_2	26.628	11.487	22.507	10.272
NCH(a)-H	3.680	1.588	3.041	1.388
NCH(b)-H	3.312	1.429	3.041	1.388
HCC=O	3.199	1.380	2.766	1.262
H(exo)	1.230	0.531	1.176	0.537
H(endo)	2.318	1.0	2.191	1.0
CH_2Me	2.046	0.883	1.839	0.839
Me	1.044	0.450	0.950	0.434

Notes to Table: Slopes are based on least-squares line fitting for data from Figures 2 and 3. Normalized values for each LSR are given relative to a value of 1.0 for the slope of the line for the signals assigned to the H(endo) resonances. Slopes were calculated based on seven experimental points up to an [LSR]:[drug] molar ratio of 0.443 for $Eu(FOD)_3$; nine points up to a molar ratio of 0.404 were used for $Eu(HFC)_3$. Correlation coefficients, R, were equal to 1.00 for all calculated line equations. See Results and Discussion.

one or (more likely) both carbonyls, based on surprisingly large LIS values for nuclei such as the HCC=O and NCH_2. Despite the considerable distance of the HCC=O and NCH_2 protons from a lanthanide bound at the NH_2 group, the LIS magnitudes for the HCC=O and NCH_2 protons are ca. 40-50% greater than that of the aryl H-2,6. In addition, larger LIS values for H(endo) vs. H(exo) also make sense if there are appreciable contributions from lanthanide bound at the carbonyls; this accounts for the observed "crossover" of the signals of this CH_2 pair. Lastly, the relatively large $\Delta\Delta\delta$ magnitudes for aryl H-3,5 and NCH_2, with added compound 3, may reflect proximity of these hydrogens to lanthanide bound at NH_2 (near aryl H-3,5) or at the carbonyls (near NCH_2); the aryl H-3,5 and NCH_2 show larger $\Delta\Delta\delta$ than other hydrogens closer to the chiral center, e.g., aryl H-2,6, H(exo), H(endo), etc.

The best resolution between signals of corresponding nuclei of enantiomers of compound 1, based on the valley height criterion, seems to be for the signals of the aryl H-3,5, for H(a) of the NCH_2, and for H(exo), with the last mentioned signal exhibiting a valley height of less than 10% of the average peak heights using a [3]:[1] molar ratio of 1.792. See Figure 5.

CONCLUSIONS

The 300 MHz ^{1}H NMR spectra of the aminoglutethimide analog, compound 1, have been studied in $CDCl_3$ solution at ambient temperatures with added achiral LSR, $Eu(FOD)_3$, compound 2, or chiral LSR, $Eu(HFC)_3$, compound 3. Relative lanthanide-induced shift magnitudes for each nucleus of compound 1 have been evaluated for both LSRs. Results are interpreted as consistent with major lanthanide complexation at the NH_2 with additional contributions from LSR bound at both carbonyls. Full ^{1}H assignments have been made based on chemical shifts, observed coupling constants, and COSY45 spectra (for spectra shifted by compound 3). With added LSR 3, enantiomeric shift differences are observable at high 3:1 ratios for all nuclei except for the NH_2, and modest potential for % e.e. determinations is apparent.

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Literature References

1. Finch N., Dziemian R., Cohen J., Steinetz B.G. The absolute configuration of the enantiomers of glutethimide and aminoglutethimide. Experientia 1975; 31(9): 1002-1003.

2. Graves P.E., Salhanick H.A. Stereoselective inhibition of aromatase by enantiomers of aminoglutethimide. Endocrinology 1979; 105: 52-57.

3. Wenzel T. J. NMR Shift Reagents. Boca Raton FL: CRC Press, 1987.

4. Eberhart S., Rothchild R. Optical purity determination and proton NMR spectral simplification with lanthanide shift reagents. Glutethimide, 3-Ethyl-3-phenyl-2,6-piperidinedione. Appl. Spectrosc. 1983; 37(3): 292-296.

5. Hatzis A., Rothchild R., Simons P. ^{1}H NMR spectral simplification with achiral and chiral lanthanide shift reagents. V. Aminoglutethimide, 3-(4-aminophenyl)-3-ethyl-2,6-piperidinedione. Anal. Chim. Acta. 1987; 194: 211-219.

6. Rothchild R., Sanders K., Venkatasubban K.S. NMR studies of drugs. Use of lanthanide shift reagents in polar solvent with Thalidomide. Spectrosc. Lett. 1993; 26(4): 597-619.

7. Avolio J., Thomson Eberhart S., Simons P., Rothchild R. Proton NMR spectral simplification with achiral and chiral lanthanide shift reagents. II. Ethosuximide, 3-ethyl-3-methyl-2,5-pyrrolidinedione, and comparisons with analogs. Spectrosc. Lett. 1986; 40(4): 531-537.

8. Reifsneider, Jr. R.J., Hoffman B.H., Venkatasubban K.S., Rothchild R., Martin R. NMR studies of drugs. Application of a chiral lanthanide shift reagent for potential direct determination of enantiomeric excess of Methsuximide. Spectrosc. Lett. 1991; 24(10): 1373-1394.

9. Rothchild R., Sanders K., Venkatasubban K.S. NMR studies of drugs. Phensuximide. ^{1}H and ^{13}C NMR methods with chiral solvating agents and lanthanide shift reagents. Spectrosc. Lett. 1993; 26(10): 1997-2015.

10. Keller L., Rothchild R. NMR studies of drugs. Applications of achiral and chiral lanthanide shift reagents to α-Ethyl-α-phenylsuccinimide. Spectrosc. Lett. 1992; 25(8): 1177-1197.

11. Myers C., Rothchild R. ^{1}H NMR spectral simplification with achiral and chiral lanthanide shift reagents. Fenimide. Direct simultaneous determination of four stereoisomers. Spectrosc. Lett. 1987; 20(10): 805-819.

12. Rowlands M.G., Bunnett M.A., Foster A.B., Jarman M., Stanek J. Analogues of aminoglutethimide based on 1-phenyl-3-azabicyclo[3.1.0]hexane-2,4-dione. Selective inhibition of aromatase activity. J. Med. Chem. 1988; 31(5): 971-976.

13. Francotte E., Wolf R.M., Lohmann D., Mueller R. Chromatographic resolution of racemates on chiral stationary phases. I. Influence of the supramolecular structure of cellulose triacetate. J. Chromatogr. 1985; 347(1): 25-37.

14. Francotte E., Cherkaoui S., Faupel M. Separation of the enantiomers of some racemic nonsteroidal aromatase inhibitors and barbiturates by capillary electrophoresis. Chirality 1993; 5(7): 516-526.

15. Stanek J., Alder À., Bellus D., Bhatnagar A.S., Häusler A., Schieweck K. Synthesis and aromatase inhibitory activity of novel 1-(4-aminophenyl)-3-azabicyclo[3.1.0]hexane- and -[3.1.1]heptane-2,4-diones. J. Med. Chem. 1991; 34(4): 1329-1334.

16. McCague R., Jarman M., Rowlands M.G., Mann J., Thickitt C.P., Clissold D.W., Neidle S., Webster G. Synthesis of the aromatase inhibitor 3-ethyl-3-(4-pyridyl)piperidine-2,6-dione and its enantiomers. J. Chem. Soc. Perkin Trans. I, 1989; 196-198.

17. Günther H. NMR Spectroscopy: Basic Principles, Concepts, and Applications in Chemistry, 2nd ed. New York: John Wiley, 1992; (a) p. 105; (b) p. 519.

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