

Application of Lanthanide Induced Shifts in Proton Magnetic Resonance Spectroscopy of Juvenile Hormones

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Induced Shifts, Juvenile Hormones

A downfield shift of proton signals up to 9 ppm is induced in the ^1H NMR spectra of juvenile hormone I (methyl 3,11-dimethyl-10,11-*cis*-epoxy-7-ethyl-2-*trans*, 6-*trans*-tridecadienoate) and III (methyl 10,11-epoxy-2-*trans*, 6-*trans*-farnesoate) by the lanthanide complex $\text{Eu}(\text{fod})_3$. A plot of induced shift vs the ratio of $[\text{LSR}]/[\text{substrate}]$ shows a good linear correlation for the range 0.3–1.0 mole ratio. Highest shifts are induced for the protons H-10 and H-2 adjacent to the two Lewis base groups of the hormone. Resolution due to induced shift decreases for the other signals with increasing distance of protons from the two coordination sites.

Since the discovery by Hinckley¹ in 1969 that an europium(III) complex produces a large isotropic chemical shift in the ^1H NMR spectrum of cholesterol, the technique of lanthanide induced shifts (LIS) has become one of the most powerful tools in proton and ^{13}C nuclear magnetic resonance spectroscopy^{2,3}. Besides their application in the quantitative use for conformational analysis of organic compounds in solution, lanthanide shift reagents (LSR) are of an outstanding help for a qualitative interpretation of the line pattern of normally clustered NMR spectra. We have made use of the europium(III) chelate $\text{Eu}(\text{fod})_3$ and obtained a good separation for a series of multiplet centres of the terpenoid juvenile hormones I (methyl 3,11-dimethyl-10,11-*cis*-epoxy-7-ethyl-2-*trans*, 6-*trans*-tridecadienoate, JH-I) and III (methyl 10,11-epoxy-2-*trans*, 6-*trans*-farnesoate, JH-III). The importance of these insect hormones has stimulated research in the biological and in the chemical field as well^{4–6}. Regarding LIS technique, the juvenile hormones should be good candidates from their two Lewis base groups with oxygen as the donor atom. With the assumption that basicity of both epoxy and methyl ester groups of JH-I and -III are of a comparable magnitude^{7,8}, separation of interfering resonances is to be expected from both ends of the molecule. $\text{Eu}(\text{fod})_3$ was chosen as LSR because it combines maximum shift capacity with minimum broadening of the shifted resonances, good solubility in chloroform with absence of interfering chelate resonances in the usual range of NMR frequencies.

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Material and Methods

Juvenile hormone I was a gift from Dr. J. Lhoste, Procida S.A., Puteaux, France. JH-III was synthesized according to Anderson *et al.*⁹. Both hormones were pure (>98%) from gas chromatographical analysis.

For ^1H NMR measurements, the Puls-Fourier-Transformation method was used at 90 MHz and 25 °C with a WH-90 instrument (Bruker-Physik, Karlsruhe). With 50 μs pulses, 50 scans per sample were accumulated. For measurement, 5 mg hormone was dissolved in 0.3 ml CDCl_3 , TMS was used as internal standard, and the LSR was a commercial preparation (Merck, Darmstadt) and was used without further purification.

Results and Discussion

The ^1H NMR spectrum of JH-I as shown in Fig. 1, has a cluster of signals near $\delta = 2.2$ from H-4, H-5, H-8, H-16 and H-17. The methyl groups H-13 and H-15 at $\delta = 1.0$ form another group of NMR resonances. A similar spectrum has JH-III, where the resonance signals from H-8, H-9 and H-15 are combined at $\delta = 2.18$ and those from H-4, H-5 and H-14 at $\delta = 1.65$ (Fig. 2). For comparison, the corresponding spectra after addition of LSR in a molar ratio near 1 are shown in the same figures. Assignment of the signals was confirmed by double resonance spectroscopy. Only in the LIS spectrum of JH-III, a definite discrimination between H-12 and H-13, the two symmetrical methyl groups, was not possible and the two signals assigned to H-12 and H-13 could also be reversed. A comparison of untreated and LIS spectra demon-



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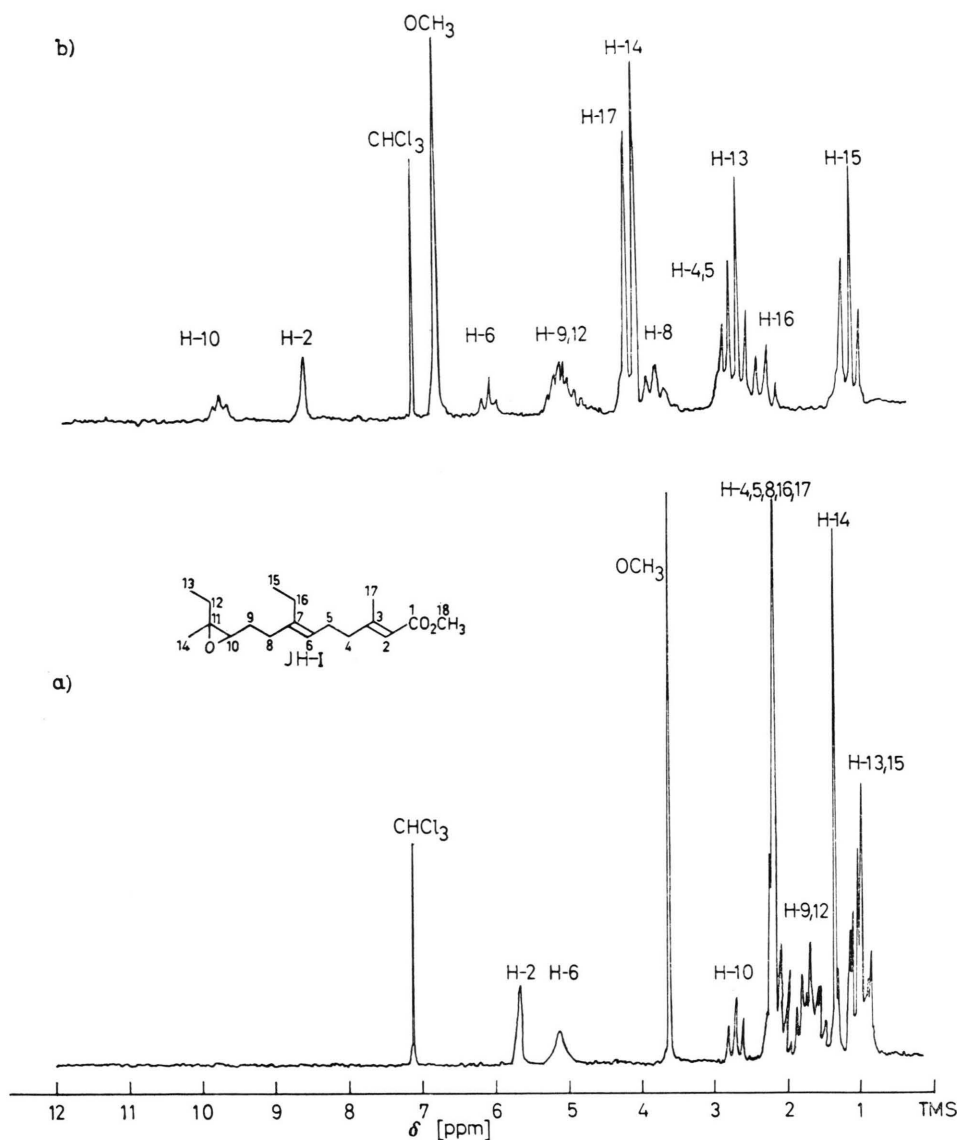


Fig. 1. 90 MHz- ^1H NMR-spectrum of 5 mg JH-I in 0.3 mol CDCl_3 with TMS as internal standard; a: without, b: with addition of LSR, $[\text{Eu}(\text{fod})_3]/[\text{JH-I}] = 0.8$. For details see Material and Methods.

strates an optimal shifting power of $\text{Eu}(\text{fod})_3$ up to 9 ppm with minimal broadening of the shifted signals. This fact indicates a predominant dipolar mechanism and excludes a remarkable contact interaction with the LSR³.

A plot of induced shift vs. the ratio of $[\text{LSR}]/[\text{substrate}]$ shows a linear correlation at least between 0.3 and 1.0 mole ratio (Fig. 3) or at even lower LSR concentration (Fig. 4). A complete separation of all signals is not achieved which is primarily the consequence of an increasing distance

of the respective protons from the two coordination sites, like H-4 and H-5 with almost no induced shift. This is in agreement with the well known fact that the influence of the paramagnetic center diminishes with increasing distance³. For JH-I, the molecular model shows for both methylene protons H-9 and H-12 the same distance and fixed angle to oxygen in the epoxy site and consequently both signals are shifted to the same extent. All the signals have a similar behaviour of shift increments also at low LSR concentrations. Signs and sizes of these in-

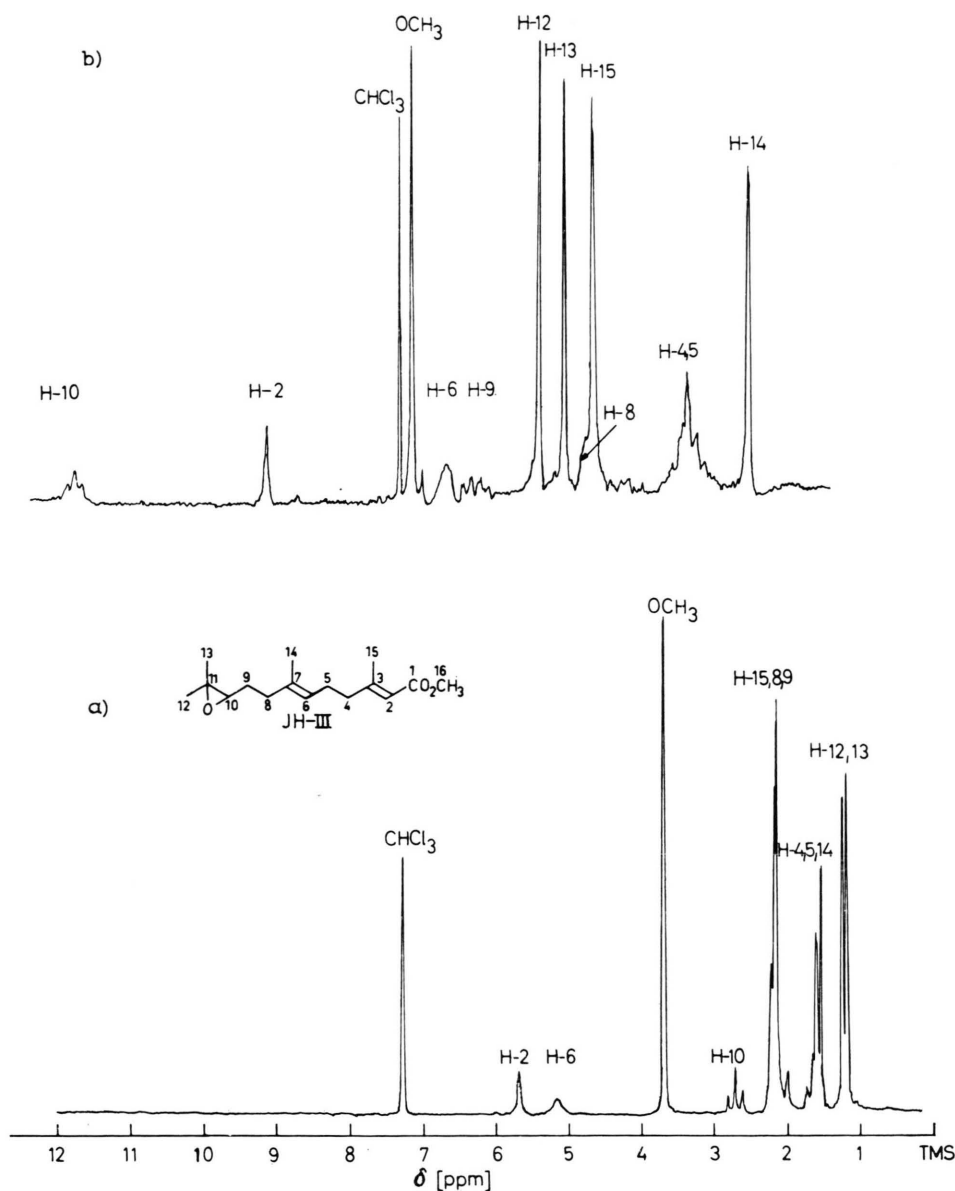


Fig. 2. 90 MHz- ^1H NMR-spectrum of 5 mg JH-III in 0.3 ml CDCl_3 with TMS as internal standard; a: without, b: with addition of LSR, $[\text{Eu}(\text{fod})_3]/[\text{JH-III}] = 1.0$. For details see Material and Methods.

duced shifts provide convincing evidence that basicities of epoxy and ester groups have about the same magnitude.

This study shows that application of LIS technique provides useful information on which structural analysis of juvenile hormones, similar terpenoid compounds, and of juvenoids can be based. The convenient and economical method allows not

only quantitative structural studies but also a simple empirical interpretation, based on distance factors only, which might be of help for routine analyses in the field of terpenes with insect hormone activity.

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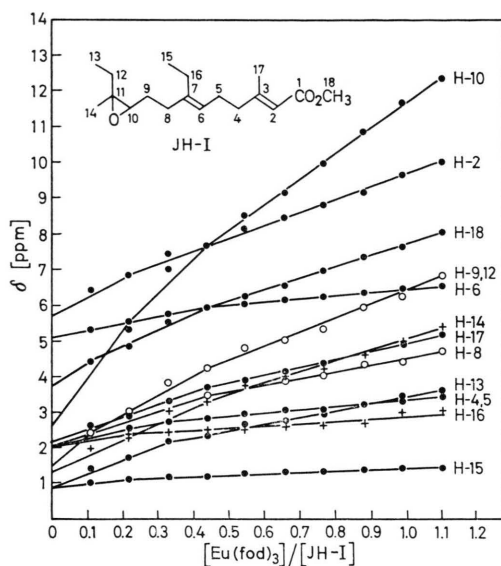


Fig. 3. Magnitude of chemical shift induced by incremental addition of $\text{Eu}(\text{fod})_3$ to a solution of 5 mg JH-I in 0.3 ml CDCl_3 .

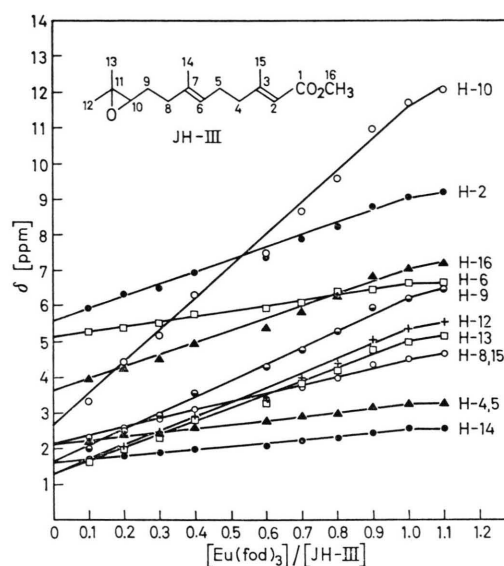


Fig. 4. Magnitude of chemical shift induced by incremental addition of $\text{Eu}(\text{fod})_3$ to a solution of 5 mg JH-III in 0.3 ml CDCl_3 .

- ¹ C. C. Hinckley, J. Amer. Chem. Soc. **91**, 5160–5162 [1969].
- ² A. F. Cockerill, G. L. Davies, R. C. Harden, and D. M. Rackham, Chem. Rev. **73**, 553–588 [1973].
- ³ O. Hofer, Topics in Stereochem. **9**, 111–197 [1976].
- ⁴ K. H. Dahm, B. M. Trost, and H. Röller, J. Amer. Chem. Soc. **89**, 5292–5294 [1967].
- ⁵ W. S. Bowers, M. J. Thompson, and E. C. Uebel, Life Sci. **4**, 2323–2331 [1965].

- ⁶ J. Menn and M. Beroza, Eds., Insect Juvenile Hormones, 339 pp., Academic Press, New York 1972.
- ⁷ P. E. Manni, G. A. Howie, B. Katz, and J. M. Cassady, J. Org. Chem. **37**, 2769–2771 [1972].
- ⁸ H. Hart and G. M. Love, Tetrahedron Lett. **7**, 625–628 [1971].
- ⁹ R. J. Anderson, C. A. Henrick, I. B. Siddall, and R. Zurflüh, J. Amer. Chem. Soc. **94**, 5379–5386 [1972].