ADP, respectively, or possibly with activation of pyruvate dehydrogenase phosphatase by Mg^{2+} and Ca^{2+}.

Regulation of the interconversion of the phosphorylated and nonphosphorylated forms of pyruvate dehydrogenase in rat adipose tissue by insulin[48,50] and in rat heart, liver, and kidney by long-chain fatty acids[51] has been reported. Insulin apparently increases the proportion of the nonphosphorylated form of pyruvate dehydrogenase, and this effect is antagonized by adrenaline and by adrenocorticotropin. Metabolic states (diabetes, starvation) associated with increased concentrations of plasma free fatty acids result in an increase in the proportion of the phosphorylated form of pyruvate dehydrogenase. It would appear that these hormonal and metabolic effects on the interconversion of the phosphorylated and nonphosphorylated forms of pyruvate dehydrogenase are indirect. It seems possible that these effects may be mediated through changes in the intramitochondrial concentrations of pyruvate, ADP, ATP, Mg^{2+}, and Ca^{2+}.

(50) S. I. Taylor, C. Mukherjee, and R. L. Jungas, *J. Biol. Chem.*, 248, 73 (1973).
(51) O. H. Wieland, C. Patzelt, and G. Löffler, *Eur. J. Biochem.*, 26, 426 (1972).

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Carbon-13 Nuclear Magnetic Resonance Spectroscopy of Naturally Occurring Substances. Alkaloids

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Research on the chemistry of organic natural products has undergone extraordinary acceleration since the advent of nuclear magnetic resonance spectroscopy in the 1950s. For the most part, this forceful tool of structure analysis has been focused on the hydrogen nucleus (pmr spectroscopy) and hence has been limited by a narrow, *ca.* 10 ppm range of spectral detail. However, recent advances in the pmr field, *e.g.*, the use of paramagnetic shift agents,[2] as well as the expansive, pioneering efforts of Lauterbur, Stothers, Grant, Roberts, and others[3] in the ^{13}C natural abundance magnetic resonance area (cmr spectroscopy), have broadened greatly the scope and utility of nmr spectroscopy for the determination of structures of natural substances.

Historically, natural product chemists have adjusted rapidly to new analytical techniques as soon as instruments have been mass produced. Since this period of development of cmr spectroscopy has arrived, broad acceptance of this new, powerful tool of analysis awaits only the accumulation of chemical shift data on compounds representative of all types of natural products. A huge data bank of the chemical shifts of alkaloids,[4] amino acids and peptides,[5] antibiotics,[6] cannabinoids,[7] carbohydrate derivatives,[8] prostaglandins,[9] steroids,[10] and terpenes[11]

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(1) (a) Public Health Service Predoctoral Fellow, 1967–1971; (b) Public Health Service Predoctoral Fellow, 1969–1972.
(2) *Inter alia*, P. V. Demarco, T. K. Elzey, R. B. Lewis, and E. Wenkert, *J. Amer. Chem. Soc.*, **92**, 5734, 5737 (1970); R. B. Lewis, Ph.D Dissertation, Indiana University, 1971.
(3) J. B. Stothers, "Carbon-13 NMR Spectroscopy," Academic Press, New York, N. Y., 1972; G. C. Levy and G. L. Nelson, "Carbon-13 Nuclear Magnetic Resonance for Organic Chemists," Wiley-Interscience, New York, N. Y., 1972; L. F. Johnson and W. C. Jankowski, "Carbon-13 NMR Spectra," Wiley-Interscience, New York, N. Y., 1972.
(4) (a) E. Wenkert, C.-J. Chang, A. O. Clouse, and D. W. Cochran, *Chem. Commun.*, 961 (1970); (b) A. Rabaron, M. Koch, M. Plat, J. Peyroux, E. Wenkert, and D. W. Cochran, *J. Amer. Chem. Soc.*, **93**, 6270 (1971); (c) E. Wenkert, D. W. Cochran, E. W. Hagaman, R. B. Lewis, and F. M. Schell, *ibid.*, **93**, 6271 (1971); (d) D. W. Cochran, Ph.D. Dissertation, Indiana University, 1971; (e) E. Wenkert, C.-J. Chang, D. W. Cochran, and R. Pellicciari, *Experientia*, **28**, 377 (1972); (f) E. Wenkert, D. W. Cochran, E. W. Hagaman, F. M. Schell, N. Neuss, A. S. Katner, P. Potier, C. Kan, M. Plat, M. Koch, H. Mehri, J. Poisson, N. Kunesch, and Y. Rolland, *J. Amer. Chem. Soc.*, **95**, 4990 (1973).
(5) (a) A. Allerhand, D. Doddrell, V. Glushko, E. Wenkert, P. J. Lawson, and F. R. N. Gurd, *J. Amer. Chem. Soc.*, **93**, 544 (1971); (b) F. R. N. Gurd, P. J. Lawson, D. W. Cochran, and E. Wenkert, *J. Biol. Chem.*, **246**, 3725 (1971); (c) P. O. Larsen, H. Sorenson, D. W. Cochran, E. W. Hagaman, and E. Wenkert, *Photochemistry*, **12**, 1731 (1973).
(6) (a) L. L. Martin, C.-J. Chang, H. G. Floss, J. A. Mabe, E. W. Hagaman, and E. Wenkert, *J. Amer. Chem. Soc.*, **94**, 8942 (1972); (b) E. Wenkert, E. W. Hagaman, and G. E. Gutowsky, *Biochem. Biophys. Res. Commun.*, **51**, 318 (1972).
(7) E. Wenkert, D. W. Cochran, F. M. Schell, R. A. Archer, and K. Matsumoto, *Experientia*, **28**, 250 (1972).
(8) E. Conway, R. D. Guthrie, S. D. Gero, G. Lukacs, A.-M. Sepulchre, E. W. Hagaman, and E. Wenkert, *Tetrahedron Lett.*, 4879 (1972).

has been acquired over the last 3 years in the Indiana University laboratories and applied to problems of structure elucidation[4a,d,e,5c,6b] and biosynthesis.[6a,11d] This Account concerns the application of ^{13}C spectroscopy to as yet cmr-unanalyzed natural products in the alkaloid field, with special attention to chemical shift data.

While the principles of ^{13}C nmr spectroscopy are the same as those of ^{1}H nmr spectroscopy, the low natural abundance of the ^{13}C isotope (1.1% as contrasted to 99.98% for the ^{1}H isotope) and its low nuclear sensitivity (1.6% of that of ^{1}H) make the acquisition of a cmr spectrum far more difficult than of a pmr spectrum. As a consequence, a larger sample size than customary for pmr spectroscopy and longer spectrometer accumulation time, if the operation is in the continuous wave mode, are needed to obtain lucid cmr spectra with high signal:noise ratio rapidly. Alternatively, data may be obtained in the Fourier transform mode.

The cmr data cited in this Account are based on ^{13}C natural abundance spectra of 0.3–1 *M* chloroform solutions[12] in spinning 13-mm o.d. tubes recorded on a spectrometer operating at 15.08 MHz with a Varian Associates DP-60 magnet working at 14 kG. In the earlier part of the present study, the spectrometer was in the continuous wave mode, while later work was based on pulsed Fourier transform spectroscopy. Currently routine high-resolution spectra run in the latter mode can be obtained on 0.5 *M* solutions in 20 min and on as low a concentration as 0.05 *M* in 20 hr.

In contrast to pmr spectra, the cmr spectrum of a complex natural product contains an uninterpretable multitude of frequently broad signals of variable intensity caused by heteronuclear coupling through one or more bonds. Two or more cmr spectra need to be run on every compound, in order to obtain data on chemical shifts and substitution patterns of individual carbon sites. A cmr spectrum executed with simultaneous irradiation of the full pmr spectral range (*i.e.*, a heteronuclear double irradiation experiment) yields a single spike for each carbon center whose high intensity reflects the coalescence of the normal multiplet and nuclear Overhauser enhancement. Such proton-decoupled spectra afford ^{13}C chemical shifts and, in the continuous wave mode, permit carbon counting.

While in a continuous wave experiment the intensity of different carbon types is similar and a signal of twice the intensity indicates two carbons with identical chemical shift, the restriction of the pulsed Fourier transform technique to specific, usually high, pulse rates leads to signals of dissimilar intensity reflecting diverse relaxation times of individual carbon sites. Since a major relaxation path involves the directly bound hydrogens, in an extreme case, especially for nonprotonated carbons, a carbon signal can be totally missing from a spectrum. In such an event the use of an intermolecular relaxing agent, *e.g.*, tris(acetylacetonato)chromium(III),[6a,13] leads to reestablishment of a spectrum of similar signal intensities.

In order to regain the information destroyed by full proton decoupling, it has become fashionable to acquire one or more types of off-resonance decoupled spectra,[3] two of which have been used extensively in the Indiana University laboratories. One, based on the single-frequency off-resonance decoupling technique (sford), by which decoupling is accomplished by irradiation with a single frequency off the pmr spectral range, produces a cmr spectrum with reduced but not eliminated coupling characteristics. It requires more signal accumulation time because of intensity reduction by coupling, even though nuclear Overhauser enhancement remains. Inasmuch as both ^{1}H and ^{13}C possess spins of ½, the individual signal multiplicity reflects the amount of hydrogen substitution at specific carbon centers by the $n + 1$ relationship familiar from pmr spectroscopy.

Since many natural products have single-frequency off-resonance decoupled spectra with uninterpretable, badly overlapping multiplets, a second off-resonance decoupling technique, which however yields more limited information, is valuable. The noise off-resonance decoupling method (nord),[11a] by which decoupling is executed by full white-noise irradiation far off the pmr spectral range, leads to the diffusion of all coupling signals and the clear retention of singlets for nonprotonated carbon sites.

In analogy with experience from pmr spectroscopy, highly substituted carbons, centers bearing electronegative groups, and multiply bonded carbon sites resonate at low fields.[3] Substitution on acyclic and five- and six-membered ring systems leads to significant shifts at the substitution site (α effect), the neighboring carbon (β effect) and the center one more carbon removed (γ effect)—a phenomenon of great importance, *inter alia*, in the determination of stereochemistry.[3]

A fundamental study of the effect of methyl substitution on the carbon shifts of the cyclohexane ring[14] and the related investigation of the piperidine nucleus[4d] permit shift evaluation of the ring systems (*vide infra*) in a predictable manner. Equatorial methyl groups impose deshielding α, β, and γ effects of 5.6, 8.9, and 0 ppm, respectively, while axial methyl functions exert similar α and β effects of 1.1 and 5.2 ppm, respectively, but a shielding γ effect of 5.4 ppm.

Occasionally cmr spectra present shifts of carbons of like substitution pattern whose differentiation by calculation of substituent parameters is at best tenuous. In such instances individual carbon recognition can be aided by specific proton–carbon decoupling, if

(9) G. Lukacs, F. Piriou, S. D. Gero, D. A. van Dorp, E. W. Hagaman, and E. Wenkert, *Tetrahedron Lett.*, 515 (1973).

(10) G. Lukacs, X. Lusinchi, E. W. Hagaman, B. L. Buckwalter, F. M. Schell, and E. Wenkert, *C. R. Acad. Sci., Ser. C*, **274**, 1458 (1972).

(11) (a) E. Wenkert, A. O. Clouse, D. W. Cochran, and D. Doddrell, *J. Amer. Chem. Soc.*, **91**, 6879 (1969); (b) E. Wenkert, A. O. Clouse, D. W. Cochran, and D. Doddrell, *Chem. Commun.*, 1433 (1969); (c) E. Wenkert and B. L. Buckwalter, *J. Amer. Chem. Soc.*, **94**, 4367 (1972); (d) J. Polonsky, Z. Baskevitch, N. Cagnoli-Bellavita, P. Ceccherelli, B. L. Buckwalter, and E. Wenkert, *ibid.*, **94**, 4369 (1972); (e) G. Lukacs, F. Khuong-Huu, C. R. Bennett, B. L. Buckwalter, and E. Wenkert, *Tetrahedron Lett.*, 3515 (1972).

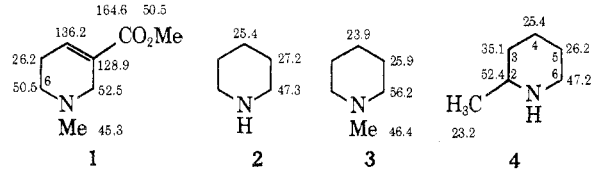
(12) All δ values on the formulas and in Table I are in parts per million downfield from TMS; $\delta(\text{TMS}) = \delta(\text{CHCl}_3) + 77.2$ ppm, the solvent being used as the internal reference.

(13) R. Freeman, K. G. R. Pachter, and G. N. LaMar, *J. Chem. Phys.*, **55**, 4586 (1971); O. A. Gansow, A. R. Burke, and W. D. Vernon, *J. Amer. Chem. Soc.*, **94**, 2550 (1972).

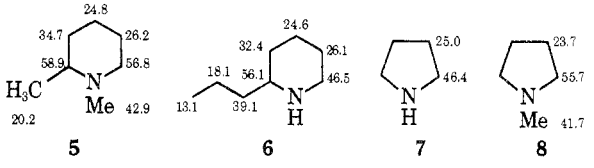
(14) D. K. Dalling and D. M. Grant, *J. Amer. Chem. Soc.*, **89**, 6612 (1967).

the pmr signal of the proton under consideration has been identified and if it exists in an isolated part of the spectrum.[3] Alternatively, specific deuterium labeling (*vide infra*) accomplishes the same task. Success of the latter technique is based on the intensity reduction or disappearance of the signal of deuterated carbons due to the reduction or loss of nuclear Overhauser enhancement and the generation of unperturbed carbon–deuterium coupling.

Application of chemical shift theory and standard decoupling techniques (*vide supra*) is sufficient for the assignment of all carbon shifts of alkaloids with gross dissimilarity of carbon sites.[4a] The analysis of the spectra of arecoline (**1**)[15] is representative. The proton-decoupled spectrum of arecoline reveals seven signals, one corresponding to two carbon centers, and its single-frequency off-resonance decoupled spectrum exhibits two singlets, one doublet, three triplets, and two quartets indicative of the presence of nonprotonated carbons, a methine, methylenes, and methyl groups, respectively. The nonprotonated carbons as well as the methyl groups are distinguished by their shift position, and the methine is unique. The methylenes fit into two categories, one upfield, and the two attached to nitrogen downfield. Differentiation of the latter two carbons is based on their dissimilar disposition to the double bond and a mild β effect of the carbomethoxy group on one of them.



1 **2** **3** **4**

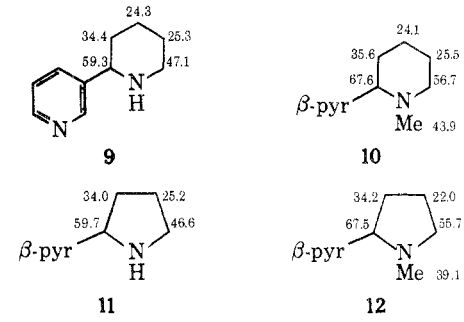


5 **6** **7** **8**

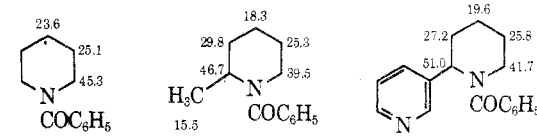
Cmr analysis of the hemlock alkaloid coniine (**6**)[16] requires prior knowledge of the shifts of piperidines **2** and **4**. The symmetry of piperidine (**2**) itself permits ready assignment of its shifts.[17] Application of them to the 2-methyl derivative (**4**), in conjunction with inspection of the spectrum of 2-methyl-2,5,5-trideuteriopiperidine[18] for differentiation of C-4 and C-5, leads then to a complete shift assignment for **4**.

Analysis of the Nicotiana bases anabasine (**9**), 1-methylanabasine (**10**), nornicotine (**11**) and nicotine (**12**)[17b] relies on interpretation of the spectra of models **4**, **5**, **7**,[17a] and **8**[17a] and knowledge of β-picoline shifts.[19]

As part of a study of piperidine bases, the spectra of *N*-benzoylpiperidines were inspected. N-Benzoylation causes mild shielding of all ring positions (*cf.* **13**). The strong shielding of the methyl group as well as C-4 and C-6 of 2-methylpiperidine on benzoylation shows the methyl function to be axial in **14**. Similar shielding of C-4 and C-6 of *N*-benzoylanabasine (**15**) suggests that the steric demand of maintaining coplanarity among at least C-2, C-6, the amide unit, and its attached benzene carbon pushes even the pyridine ring into an axial conformation.[20] Thus cmr spectroscopy appears to be a powerful tool of conformational analysis.



9 **10** **11** **12**



13 **14** **15**

The symmetry-induced, structural simplicity of most tropane alkaloids, their alkamines, and derivatives render this group of substances an easy object for cmr spectral determination and enables the latter to yield information on subtle aspects of structure more efficiently than most present-day methods of instrumental analysis. Nortropane (**16**), the basic skeleton of the alkaloid family, reveals four cmr peaks of which the farthest downfield corresponds to the aminomethines and the farthest upfield the piperidino γ carbon. Evaluation of the remaining methylene signals depends on comparison with the β-methylene shift values of piperidine (**2**) and pyrrolidine (**7**) and expected axial α-alkyl substituent effects.[3]

The chemical shifts of tropane (**17**) and its oxygenated derivatives, including the alkaloids atropine (**22**) and tropacocaine (**24**), are depicted on the formulas. The weak shielding of C-2 and C-4 and strong upfield shift of C-6 and C-7 indicate the *N*-methyl group to be oriented equatorially in the piperidine ring,[21] while the mild effect of the change of C-3 substituent stereochemistry in the epimer pairs **20**–**23** and **21**–**24** on the shifts of the ethano bridge and C-3 indicates a diminution of the difference of equatorial and axial substituent behavior, *i.e.*, flattening of the piperidine ring, in consonance with the known molecular shape of pseudotropine (**23**) in the solid state as deduced from X-ray crystallography.[22]

The lack of symmetry of tropidine (**25**) makes its cmr analysis difficult. However, a cmr effect first ob-

(15) In carbon tetrachloride solution; $\delta(\text{TMS}) = \delta(\text{CCl}_4) + 95.9$ ppm.

(16) In dioxane soution; $\delta(\text{TMS}) = \delta(\text{dioxane}) + 66.3$ ppm.

(17) (a) G. E. Maciel and G. B. Savitsky, *J. Phys. Chem.*, **69**, 3925 (1965); (b) W. O. Crain, Jr., W. C. Wildman, and J. D. Roberts, *J. Amer. Chem. Soc.*, **93**, 990 (1971); (c) I. Morishima, K. Okada, T. Yonezawa, and K. Goto, *ibid.*, **93**, 3922 (1971).

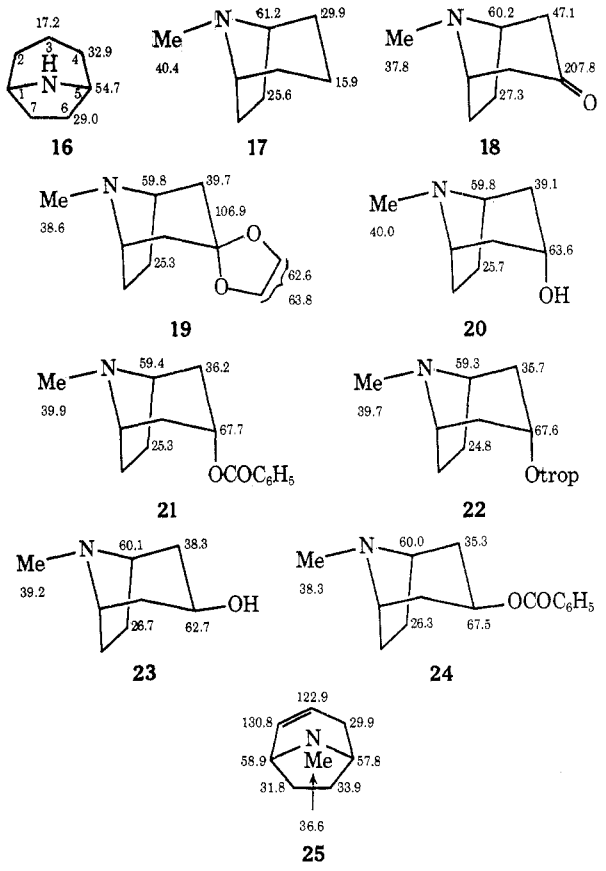
(18) The deuterated piperidine was prepared by consecutive base-induced deuteration of 2-methylcyclopentanone, Beckmann rearrangement of its trideuterio ketoxime under the influence of deuteriopolyphosphoric acid, and lithium aluminum hydride reduction of the resultant piperidone.

(19) P. C. Lauterbur, *J. Chem. Phys.*, **43**, 360 (1965).

(20) F. Johnson, *Chem. Rev.*, **68**, 375 (1968).

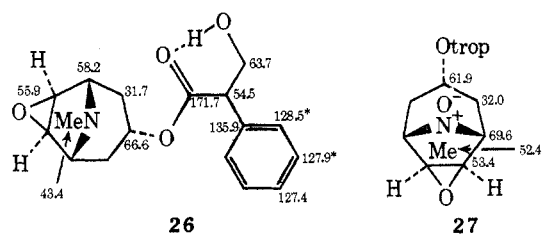
(21) Tropine (**20**) methobromide in aqueous methanol shows two distinct *N*-methyl signals, axial and equatorial peaks with respect to the piperidine ring, at 43.4 and 50.2 ppm, respectively (E. Wenkert, D. W. Cochran, and F. M. Schell, unpublished observations).

(22) H. Schenk, C. H. MacGillavry, S. Skolnik, and J. Laan, *Acta Crystallogr.*, **23**, 423 (1967).



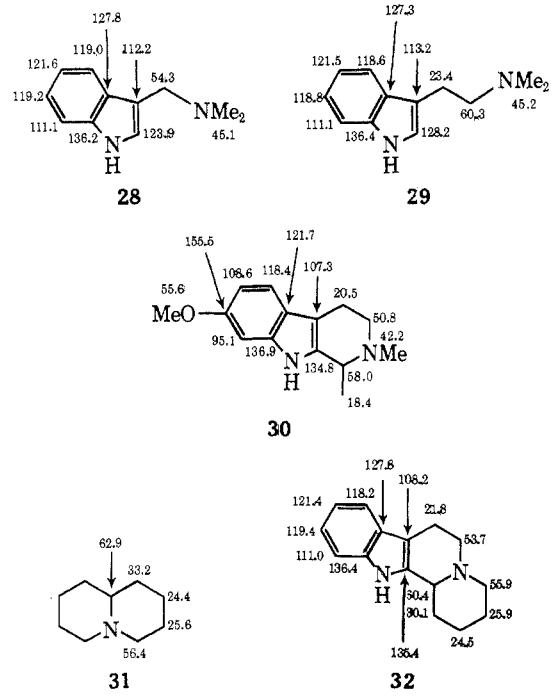
16 17 18 19 20 21 22 23 24 25

served among piperideines aids in the shift assignment. Double bonds in six-membered rings have a shielding effect on the endocyclic homoallyl carbons,[4d,f] *e.g.*, $\Delta\delta^{C\text{-}6}$ = 5.0 ppm for arecoline (1) and its dihydro derivative. If this effect is applicable to seven-membered rings, the methines as well as ethano bridge carbons of tropidine (**25**) can be differentiated from each other. Olefinic C-2 appears at lower field than C-3 in view of greater substitution at C-1 than at C-4. Comparison of the cmr spectra of the alkaloids scopolamine (**26**) and scopolamine oxide (**27**) permits evaluation of the shifts of their oxidotropinol units,[23] while analysis of their tropic acid fragments (*cf.* also **22**) is based on simple chemical shift theory.[3,24] Amine oxides prove to be excellent alkaloid derivatives for cmr assay in view of strong chemical shift changes and the ease of their reductive reversion to the natural bases.



26 27

Analysis of the complex indole alkaloids of the tetrahydrocarboline type is accomplished most easily by an initial chemical shift evaluation of simple indole bases. In view of the availability of literature values of aromatic shifts of indole models,[25] the carbon shifts of the alkaloids gramine (**28**) and *N,N*-dimethyltryptamine (**29**) and the alkaloid model N_b-methyltetrahydroharmine (**30**) can be assigned. Furthermore, use of these shifts and those of 1,2-dimethylpiperidine (**5**) and quinolizidine (**31**), obtained with the aid of a cmr spectrum of its 3,3-dideuterio derivative,[26] permits interpretation of the spectrum of the *Dracontomelum mangiferum* base **32**.[27] Comparison of the aminomethylene shifts of **31** and **32** shows C-21 of the latter being nearly identical with the related shift of quinolizidine (**31**) and C-5 differing by the endocyclic homoallyl effect.[4d,f]



28 29 30 31 32

The cmr analysis of base **32** lays the foundation for similar studies of the indole alkaloids of the yohimboid, ajmalicinoid, and corynantheioid types.[4d] Signal assignment of rings A, B, and C of the natural bases corynantheine (**33**), dihydrocorynantheine (**34**), and corynantheidine (**35**) is derived directly from the data of **32**. Acquisition of the δ values of the β-methoxyacrylate moiety, C-18 and C-19 of **33** and C-18 of **34** and **35**, is attained by application of standard chemical shift theory. The two remaining methylenes of corynantheine (**33**) are differentiated by one being an aminomethylene group. Two of the three remaining methylenes of dihydrocorynantheine (**34**) show similar chemical shifts. Inasmuch as **34** possesses an equatorial ethyl group and **35** has its ethyl function in an axial conformation, the axial C-19 of the latter should exert strong β and γ effects on C-21 and C-14, respectively. These manifestations are observed and duplicated in the model aldehyde **36**. The difference of C-20 substituents in **33** and **34** distinguishes the C-15 and C-20 shifts in these alkaloids, while the variation of the C-15 substituent in

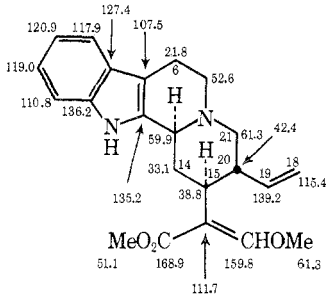
(23) Assessment of the importance of steric, dipolar, and conformational effects as possible causes for the anomalous δ values of especially C-2, C-4, and the *N*-methyl group will have to await future work.

(24) Starred δ values on the formulas indicate that they may be interchanged.

(25) R. G. Parker and J. D. Roberts, *J. Org. Chem.*, **35**, 966 (1970).

(26) This substance was prepared by base-catalyzed deuteration of 4-quinolizidone, followed by lithium aluminum hydride reduction.

(27) E. Wenkert, R. A. Massy-Westropp, and R. G. Lewis, *J. Amer. Chem. Soc.*, **84**, 3732 (1962); E. Wenkert and B. Wickberg, *ibid.*, **84**, 4914 (1962); E. Wenkert, K. G. Dave, and F. Haglid, *ibid.*, **87**, 5461 (1965); S. R. Johns, J. A. Lamberton, and J. L. Occolowitz, *Aust. J. Chem.*, **19**, 1951 (1966).



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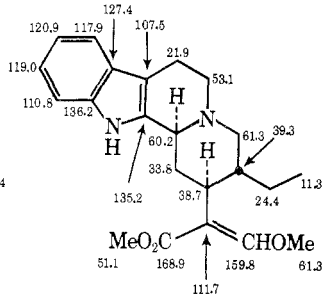

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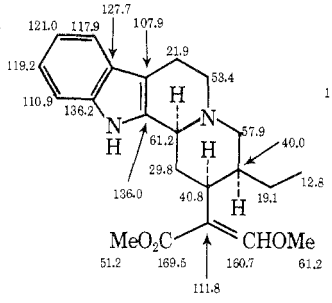

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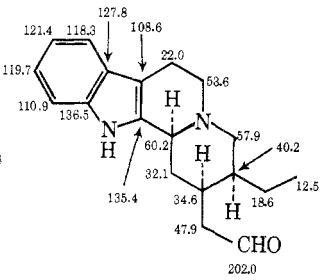
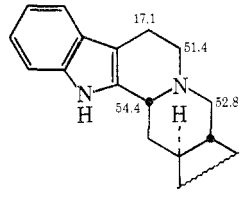


36

35 and **36** differentiates the same methines in this base pair.

As a cmr study of the four different stereochemical types of yohimbines indicated,[4b] the pseudo configuration (**37**), held rigidly in *cis*-quinolizideine form, exhibits ring C and D carbon shifts greatly different from those of the other three stereostructures, *e.g.*, the *normal* configuration (**38**), kept in a *trans*-quinolizideine form. Comparison of these data with those of all the above quinolizideines shows the latter to be trans substances.



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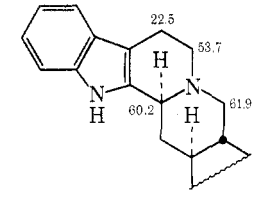

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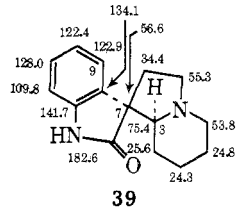
Previous chemical shift information on oxindoles[4a] and the data on 1,2-disubstituted piperidines and pyrrolidines permit complete analysis of the stereoisomeric oxindole alkaloid models **39** and **40**[28] as well as indolizidine (**41**).[29] A stereochemically significant fact emanates from this study. The chemical shift values of C-3 and C-9 are strongly diagnostic of the configuration of the spiro carbon. This observation is most useful in the cmr analysis of the alkaloids rhyncophylline (**43**) and isorhyncophylline (**44**). Full shift assignment of the natural bases and the degradation product rhyncophyllal (**42**) is dependent on the δ values of dihydrocorynantheine (**34**) and models **39** and **40**.

(28) The substances **39**, mp 195–196°, and **40**, mp 146–147°, were prepared by treatment of the alkaloid **32** with *tert*-butyl hypochlorite and of the resultant β-chloroindolenines consecutively with methanolic hydroxide and aqueous acid. Their stereochemistry was determined by equilibrium studies of each isomer: refluxing a pyridine solution yielded exclusively **40**, while refluxing a 10% aqueous acetic acid solution gave a 2.3:1 mixture of **39** and **40**. This work (E. Wenkert and J. S. Bindra, unpublished observations) preceded a recent report on these compounds [A. H. Gaskell, H.-E. Radunz, and E. Winterfeldt, *Tetrahedron*, **26**, 5353 (1970)].

(29) Interpretation of the spectrum of 5,6,7,8,8a-pentadeuterioindolizidine, prepared by catalytic deuteration of 1,2,3-trihydroindolizinium bromide, verified the analysis.



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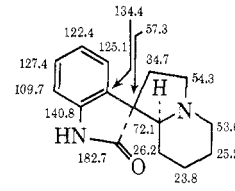

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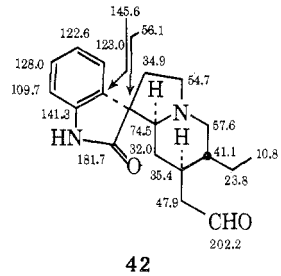

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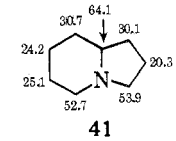

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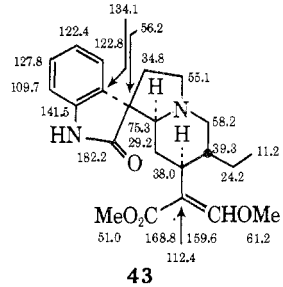

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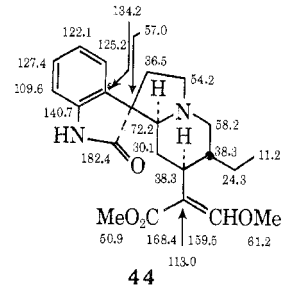

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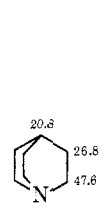
The challenge of the structurally complex Cinchona bases is the analysis of their substituted quinuclidine and quinoline units. Quinuclidine (**45**)[17c] itself and its oxide (**46**) serve as models for the former moiety, while the methoxyquinoline portion of the Cinchona alkaloids quinine (**47**) and quinidine (**48**) is diagnosed with the help of quinoline[30] and aromatic substituent parameters derived from anisole[31] and γ-picoline.[19] Chemical shift data (Table I) of quinine N_b-oxide (**47a**), 18,19-dihydroquinine (**47b**), and 18,19-dihydroquinine N_b-oxide (**47c**) as well as the same derivatives of quinidine (**48a**, **48b**, **48c**, respectively) act as further aids.



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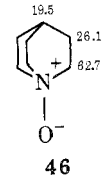

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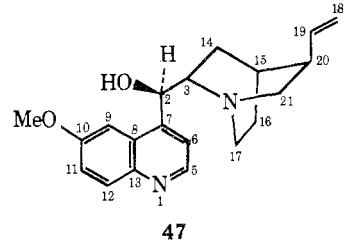

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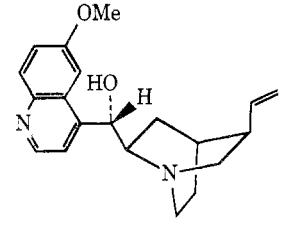


48

The nonaromatic fragment of quinine (**47**) and, hence, of the other Cinchona systems is analyzed as

(30) R. J. Pugmire, D. M. Grant, M. J. Robins, and R. K. Robins, *J. Amer. Chem. Soc.*, **91**, 6381 (1969).

(31) H. Spiesecke and W. G. Schneider, *J. Chem. Phys.*, **35**, 731 (1961); P. C. Lauterbur, *J. Amer. Chem. Soc.*, **83**, 1846 (1961); F. S. Dhami and J. B. Stothers, *Can. J. Chem.*, **44**, 2855 (1966).

Table I
^{13}C Chemical Shifts[12]

	47	47a	47b	47c	48	48a	48b	48c
C-(2)	71.4	73.2	71.2	73.2	71.7	72.9	71.5	72.7
C-(3)	60.0	62.9	59.6	63.2	60.0	62.8	59.8	62.6
C-(5)	147.0	147.2	146.8	147.1	147.3	147.4	147.2	147.6
C-(6)	121.2	122.0	120.7	121.9	121.3	122.1	121.4	121.7
C-(7)	148.6	147.2	148.7	147.2	148.3	147.4	148.7	147.6
C-(8)	126.6	126.0	126.4	125.9	126.8	125.8	126.6	125.7
C-(9)	101.7	100.6	101.6	100.8	101.8	100.4	101.6	100.0
C-(10)	158.5	157.8	157.4	157.8	157.7	157.7	157.6	157.4
C-(11)	118.5	119.0	118.2	119.1	118.5	119.2	118.6	119.1
C-(12)	130.8	131.0	130.6	131.1	131.3	130.9	131.0	130.8
C-(13)	143.9	143.9	143.6	143.8	144.0	143.7	144.0	143.5
C-(14)	20.7	20.2	20.6	20.2	21.2	20.2	20.6	19.8
C-(15)	27.7	27.2	25.2	25.2	28.4	27.7	26.0	25.3
C-(16)	27.7	27.2	28.0	27.7	26.7	26.6	26.7	27.0
C-(17)	43.1	58.9	42.9	59.0	49.5[a]	63.7[a]	51.1[a]	65.5
C-(18)	113.7	116.4	11.6	11.4	114.5	111.6	12.0	11.6
C-(19)	141.7	138.3	27.3	27.3	140.6	147.4	25.1	24.9
C-(20)	39.9	40.9	37.3	39.3	40.1	41.3	37.4	38.8
C-(21)	56.9	70.8	58.2	73.2	50.0[a]	65.4[a]	50.1[a]	65.5
OMe	55.5	54.9	55.3	55.1	55.6	54.6	55.5	54.6

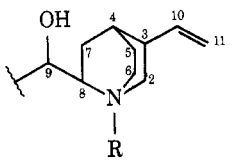
[a] Values within any vertical column may be reversed.

follows. Carbons 2 and 3 are expected farthest downfield, while the remaining methines C-15 and C-20 are recognized from the quinuclidine (45) shifts and substituent parameters.[3] The downfield aminomethylenes C-17 and C-21 can be distinguished by the anticipation of the former being shielded by the C-3 substituent and the latter being deshielded by the vinyl group in quinine (47) as well as C-17 being unaffected and C-21 being shielded by the quinolylcarbinol moiety and deshielded by the vinyl group in quinidine (48). Similarly, the methylenes C-14 and C-16 can be predicted to differ by the latter being unaffected by substituents in both quinine (47) and quinidine (48) and thus being a normal quinuclidino β-methylene.[32]

The above evaluation of the ^{13}C chemical shifts of a limited group of organic natural products and of their significance in discerning structural detail shows that cmr spectroscopy is an invaluable aid in organochemical and biochemical research.[33]

The authors acknowledge with gratitude gifts of alkaloid samples from Drs. L. Dorfman (Ciba Pharmaceutical Co.), G. Fodor (University of West Virginia), E. J. Fornefeld (Eli Lilly and Co.), R. Goutarel (I.C.S.N., Gif-sur-Yvette, France), J. A. Lamberton (C.S.I.R.O., Melbourne, Australia), and N. Neuss (Eli Lilly and Co), the able technical assistance of Mr. L. D. Kwart in the preparation of synthetic intermediates, and the financial support of the National Science Foundation and the Petroleum Research Fund (administered by the American Chemical Society) to Professor A. Allerhand for the development and construction of the departmental Fourier transform cmr equipment. Special thanks are extended to Mr. A. O. Clouse whose extraordinary ingenuity and persevering research drive in electronics and engineering are responsible for the establishment of an outstanding nmr facility at Indiana University and for enabling the work described in this Account to proceed. Finally, E. W. wishes to express his personal thanks to his former student, Dr. David Doddrell, for first introducing him to cmr spectroscopy.

(32) A cmr analysis of the nonaromatic part of quinine (47) and its dimethiodide has been presented without explanation by Roberts.[17b] Since the structural formula representing these substances was drawn incorrectly and left unnumbered, the accompanying table of chemical shifts is uninterpretable. Comparison of the cited data with the δ values for 47 in Table I indicates that the numbering system in formula i had been used. If this be so, the chemical shifts of C-5 and C-7 [*i.e.*, C-16 and C-14 in 47, respectively] are at variance with the above assignments.



i

(33) **Added June 28, 1973.** Since completion of the cmr studies depicted in this Account and its submittal for publication the following articles presenting identical or related data have appeared: piperidines and pyrrolidines: ref 3; G. Ellis and R. G. Jones, *J. Chem. Soc., Perkin Trans. 2*, 437 (1972); A. J. Jones, A. F. Casey, and K. M. J. McErlane, *Tetrahedron Lett.*, 1727 (1972); A. J. Jones and M. M. A. Hassan, *J. Org. Chem.*, **37**, 2332 (1972); D. Wendisch, H. Feltkamp, and M. von V. Scheidegger, *Org. Mag. Res.*, **5**, 129 (1973); A. J. Jones, A. F. Casey, and K. M. J. McErlane, *Can. J. Chem.*, **51**, 1782 (1973); A. J. Jones, C. P. Beeman, A. F. Casey, and K. M. J. McErlane, *ibid.*, **51**, 1790 (1973); *Dracontomelum* base **32**: ref 3; G. W. Gribble, R. B. Nelson, G. C. Levy and G. L. Nelson, *J. Chem. Soc., Chem. Commun.*, 703 (1972) [the signals of C-6, C-15, and C-20 were misassigned]; quinuclidine: J. C. Coll, D. R. Crist, M. d. C. G. Barrio, and N. J. Leonard, *J. Amer. Chem. Soc.*, **94**, 7092 (1972); G. Van Binst and D. Tourwe, *Org. Mag. Res.*, **4**, 625 (1972).