with only an 11% yield of indene (**6**). Since the desired naphthol requires CO insertion, the effect of CO pressure was tested. With a small positive pressure of CO (balloon), no significant change in rate or ratio of naphthol to indene was noted. At 4.5 atm of CO, the reaction is strongly inhibited; in addition, no naphthol was detected while indenes formed as usual after extended time.

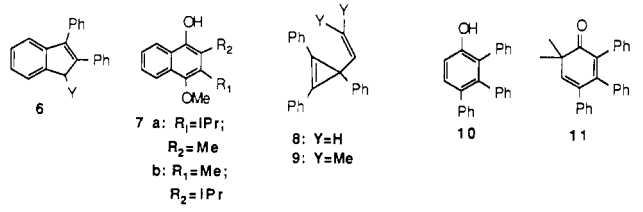
Triphenylcyclopropene was used to demonstrate catalytic activity by $Mo(CO)_6$. In each case, while the same reaction conditions (dioxane, reflux, 1.1 atm of CO) and the same concentration of 1,2,3-triphenylcyclopropene were maintained, the amount of $Mo(CO)_6$ was decreased and the isolated yield of naphthol **2b** was monitored:

$Mo(CO)_6$ (mol equiv)	naphthol **2b** (yield)
1.0 (50 min; no CO)	78%
0.33 (12 h)	73%
0.10 (2.5 h)	63%
0.07 (22 h)	46%

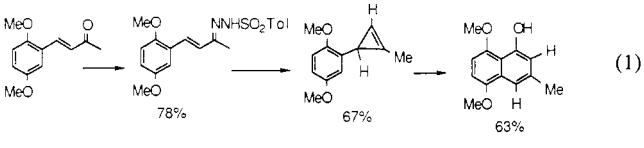
The data demonstrate six or so turnovers of $Mo(CO)_6$ but rather low rates.

A crucial issue in the naphthol synthesis by the alkyne–carbene complex cycloaddition is the regioselectivity in coupling to the alkyne. We have examined this feature of the cyclopropene rearrangements and find a somewhat lower degree of selectivity and *opposite* orientation (Table II, entries 2–7). For example, reaction of isopropylmethylacetylene with carbene complex **1** produces naphthols **7b** and **7a** in the ratio 83:17 (compare Table II, entries 3 and 4).

The special examples **8** and **9** show preferential migration of the vinyl group, leading to monocyclic products **10** (62% yield) and **11** (32% yield), respectively.[20] Substrate **8** is among the most reactive of the cyclopropenes studied, giving complete conversion with $Mo(CO)_6$ after 15 min in 1,2-dimethoxyethane at reflux (83 °C).



The rearrangements reported here provide suggestive evidence for the key intermediates proposed in the alkyne–carbene complex cycloaddition, but no intermediates have been detected. Overall, the metal carbonyl promoted cyclopropene rearrangement has several potential virtues as a preparative method for naphthols: catalytic use of the metal, versatility in the substituents on the naphthol, and several general procedures available to prepare the cyclopropene starting material. In a favorable case illustrated in eq 1, the overall procedure is quite efficient, starting from the aldol product of acetone and 2,5-dimethoxybenzaldehyde.[21]



(1)

(18) Indenes are common products from thermal and Lewis acid promoted rearrangements of phenylcyclopropenes.[19] We have demonstrated that the indene formation reported is catalyzed by the metal carbonyl and is not a direct thermal rearrangement.

(19) (a) Breslow, R. In *Molecular Rearrangements*; de Mayo, P., Ed.; *Interscience*: New York, 1963; Part 1, p 257. (b) McCullough, J. *Can. J. Chem.* **1968**, *46*, 43. (c) Battiste, A.; Halton, B.; Grubbs, R. H. *Chem. Commun.* **1967**, 907. (d) Walker, J. A.; Orchin, M. *Ibid.* **1968**, 1239.

(20) Cho and Liebeskind have investigated rhodium-catalyzed rearrangement of 2-acyl- and 3-vinylcyclopropenes and report phenol formation from the latter. We thank Professor Liebeskind for communicating the results prior to publication.

(21) We wish to acknowledge support from the National Institutes of Health in the form of postdoctoral fellowships to M. Steigerwald and S. Ho and a research grant (CA26727) to M. F. Semmelhack.

Geminal Dimethyl Stereochemistry in the Enzymatic Cyclization of Geranyl Pyrophosphate to (+)- and (−)-α-Pinene

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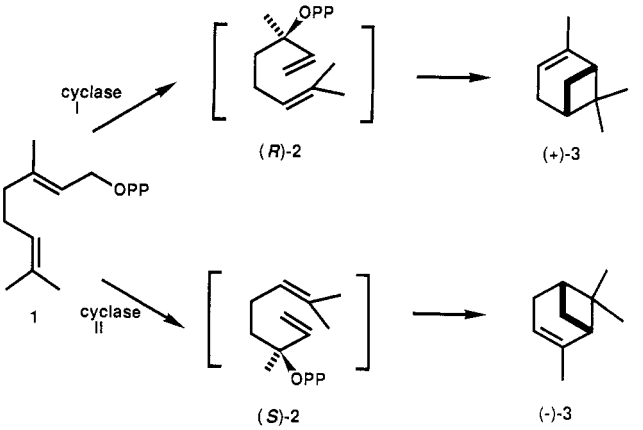
Although a geminal dimethyl bridge is a common structural characteristic among bicyclic monoterpenes, the stereochemistry of the cyclizations which form these prochiral centers is presently unknown. According to current concepts,[2,3] the enantiomeric bridged bicyclic monoterpenes[4,5] are biosynthesized from geranyl pyrophosphate (**1**) via enzyme-catalyzed isomerization[6] to (*R*)- or (*S*)-linalyl pyrophosphate (**2**) followed by anti-S_N' cyclization to C-6 and electrophilic attack of C-7 upon the endocyclic double bond of a transient α-terpinyl carbocation. Previous work[7] has achieved separation of two pinene cyclase activities (I and II) from immature leaves of the common sage plant (*Salvia officinalis*) which catalyze the stereospecific conversion of geranyl pyrophosphate to (+)- and (−)-α-pinene (**3**), respectively. The stereochemistry of (+)-pinene cyclase I and (−)-pinene cyclase II with respect to C-1 of geranyl pyrophosphate (retention) and the configuration of the preferred linalyl pyrophosphate substrate ((*R*)-**2** → (+)-**3**, (*S*)-**2** → (−)-**3**)[2b,8] are consistent with anti-S_N' cyclizations from enantiomeric endo conformations of the presumed tertiary pyrophosphate intermediate.[2,3,9] We wish to report the results of an investigation to elucidate the stereochemical course of these cyclizations at the geminal dimethyl position of the α-pinene enantiomers.



(6*E*)-[8-^{3}H]Geraniol (9.1 mCi, 62 mCi/mmol) was synthesized from (6*E*)-8-hydroxygeranyl benzyl ether[10] in five steps (36%

(1) National Institutes of Health trainee, 1984–1987 (PHS 5 T32 GM 07283).

(2) (a) Croteau, R. In *Biogeneration of Aromas*; Parliment, T. H., Croteau, R., Eds., American Chemical Society: Washington, DC, 1986; pp 134–156. (b) Croteau, R. In *Biochemistry of Plant Lipids: Structure and Function*; Stumpf, P. K., Ed.; Plenum: New York, 1987; pp 11–18.

(3) Cane, D. E. *Acc. Chem. Res.* **1985**, *18*, 220–226.

(4) (+)- and (−)-bornyl pyrophosphate: (a) Croteau, R.; Felton, N. M.; Wheeler, C. J. *J. Biol. Chem.* **1985**, *260*, 5956–5962. (b) Croteau, R.; Satterwhite, D. M.; Cane, D. E.; Chang, C. C. *J. Biol. Chem.* **1986**, *261*, 13438–13445.

(5) (−)-*endo*-Fenchol: Satterwhite, D. M.; Wheeler, C. J.; Croteau, R. *J. Biol. Chem.* **1985**, *260*, 13901–13908.

(6) A suprafacial PP migration is assumed by analogy with cyclonerodiol (a) and linalool (b) biosynthesis: (a) Cane, D. E.; Iyengar, R.; Shiao, M.-S. *J. Am. Chem. Soc.* **1981**, *103*, 914–931. (b) Godtfredsen, S. E., Ph.D. Thesis, ETH Zurich, No. 6243.

(7) Gambliel, H.; Croteau, R. *J. Biol. Chem.* **1984**, *259*, 740–748.

(8) Croteau, R.; Satterwhite, D. M., unpublished results.

(9) Godtfredsen, S.; Obrecht, J. P.; Arigoni, D. *Chimia* **1977**, *31*, 62–63. See also ref 6b.

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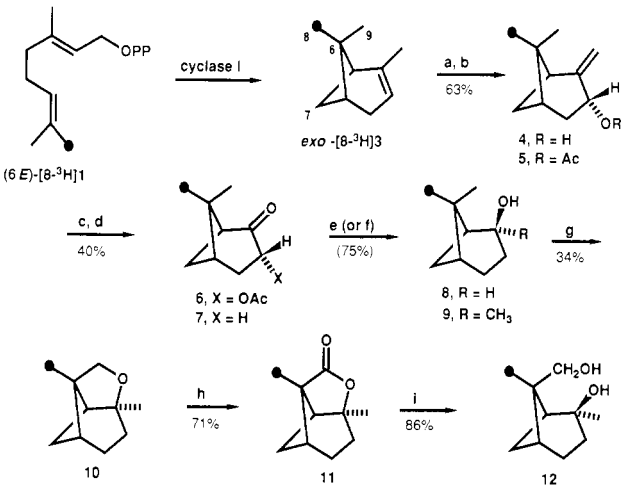
Scheme I[a]

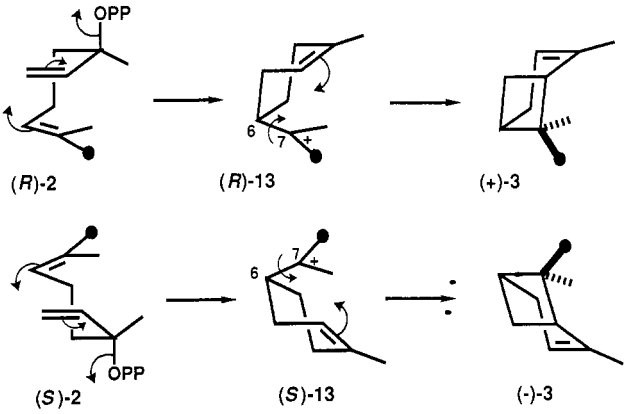


[a] Reagents: (a) $ArCO_3H$, $LiNEt_2$; (b) Ac_2O, pyr; (c) O_3, MeOH; (d) Li, NH_3, −78 °C; (e) $NaBH_4$, THF, H_2O; (f) CH_3Li, ether, two recycles; (g) HgO, Br_2, $h\nu$, pentane; (h) $ZnCr_2O_7 \cdot 3H_2O$, CH_2Cl_2; (i) $LiAlH_4$, ether.

radiochemical yield),[11] assayed for specific radioactivity as its recrystallized 3,5-dinitrobenzoate (mp 62–62.5 °C), and converted to (6*E*)-[8-^{3}H]geranyl pyrophosphate by the modified Cramer–Böhm[12] procedure. The pinene cyclases were separated on large scale from the 140 000-g supernatant of sage leaf homogenates by concentration and gel permeation chromatography on Sephacryl S-200 according to previously described procedures and assays.[7,13] Separate incubations[14] of the [8-^{3}H]geranyl pyrophosphate with each of the purified cyclases afforded mixtures of isomeric monoterpene hydrocarbons (14–29% radiochemical conversion)[15] from which [^{3}H]-α-pinene was isolated by preparative TLC on silver nitrate impregnated silica gel after addition of ca. 100 mg of racemic carrier.[16]

(+)-[^{3}H]-α-Pinene from cyclase I was further diluted with (±)-α-pinene (2 g) and converted to *trans*-[^{3}H]pinocarveol (**4**)[17,18] via epoxidation and β-elimination.[19] Ozonolysis of the corresponding acetate (**5**) followed by lithium ammonia reduction of the resulting α-acetoxy ketone (**6**) afforded [^{3}H]nopinone (**7**). The tritium content was quantitatively determined at this stage by reduction to [^{3}H]nopinol (**8**)[20] and crystallization to constant specific activity (0.037 ± 0.002 μCi/mmol after correction for a second dilution with (±)-**7**).[21] Reaction of [^{3}H]nopinone with methyllithium in ether gave *trans*-[^{3}H]-2-pinanol (**9**) which was oxidized to tricyclic ether **10** according to the procedure of Gibson and Erman.[22] Oxidation of **10** to lactone **11** with "zinc chromate trihydrate"[23] followed by reduction with lithium aluminum hydride gave the known diol **12**.[22] The specific activity of (±)-**12** (mp 57.5–58 °C) remained at 0.039 ± 0.001 μCi/mmol during three crystallizations from hexane.

The complete retention of radioactivity establishes that the tritium of the substrate resides entirely in the exo methyl group of the product as shown in Scheme I.[24] The same outcome resulted when (−)-[^{3}H]-α-pinene (1.0 μCi)[25] from cyclase II was diluted with (−)-α-pinene carrier and converted to (+)-[^{3}H]nopinone (0.026 ± 0.002 μCi/mmol)[25] and the corresponding (−)-[^{3}H]diol (mp 84.5–85.0 °C, 0.023 ± 0.002 μCi/mmol). It is therefore clear that both enzymatic cyclizations are stereospecific to the limits of the measurements (≥90–95%) and that the *E* methyl of the substrate becomes the exo methyl group in both (+)- and (−)-α-pinene. Consequently, the initial anti,endo cyclizations of (*R*)- and (*S*)-linalyl pyrophosphate are evidently followed by enantiomeric, least-motion (30° vs. 150° rotation about the exocyclic C_6–C_7 bond) cyclizations of the boat-like α-terpinyl intermediates (**13**) to form the cyclobutane rings of these monoterpenes.



The methyl group correlation in α-pinene biosynthesis is in harmony with the exo stereochemistry of the more common α- and β-bergamotenes,[26] if it is assumed that these bicyclo[3.1.1]heptane-based sesquiterpenes are derived from (*E*,*E*)-

(10) Coates, R. M.; Ley, D. A.; Cavender, P. L. *J. Org. Chem.* **1978**, *43*, 4915–4922.

(11) (a) $(COCl)_2$, Me_2SO, CH_2Cl_2, −60 °C, Et_3N; (b) $NaBT_4$, EtOH; (c) NCS, Me_2S, CH_2Cl_2, −40 to 25 °C; (d) $LiBEt_3H$, THF, 0 to 25 °C; (e) Li, NH_3, THF, −78 °C. The regiospecificity of this reaction sequence was established by synthesis of (6*Z*)-[8-^{13}C]geraniol in a similar manner.

(12) Reaction procedure: (a) Cramer, F.; Böhm, W. *Angew. Chem.* **1959**, *71*, 775. (b) Cramer, F.; Rittersdorf, W. *Tetrahedron* **1967**, *23*, 3015–3021. (c) Cornforth, R. H.; Popjak, G. *Methods Enzymol.* **1969**, *15*, 359–390. (d) Ion-exchange separation: Sofer, S. S.; Rilling, H. C. *J. Lipid Res.* **1969**, *10*, 183–187. (e) Amberlite column: Fall, R. R.; West, C. A. *J. Biol. Chem.* **1971**, *246*, 6913–6928.

(13) Gambiel, H.; Croteau, R. *J. Biol. Chem.* **1982**, *257*, 2335–2342.

(14) Incubation conditions: 5-mL portion of cyclase I (10 mM Na_2HPO_4, 1 mM Na ascorbate, 0.5 mM dithiothreitol, 10% glycerol, pH 6.2), 10 mM $MgCl_2$, 25 μM [8-^{3}H]geranyl pyrophosphate, 2 mL of pentane overlay, 30 °C, 16 h; cyclase II, similar except 200 mM HEPES buffer at pH 7.2. The hydrocarbon products were isolated by extraction with pentane and passage through silica gel.

(15) α-Pinene, β-pinene (only from cyclase (II), camphene, limonene, and myrcene. [^{3}H]-α-Pinene comprised 25–30% of the [^{3}H]monoterpenes obtained from cyclase I and 15–20% of those from cyclase II.

(16) The enantiomeric purity of the [^{3}H]-α-pinene products was determined by conversion to isopinocamphone ((a) BH_3, THF; H_2O_2, OH^-. (b) PCC, CH_2Cl_2), formation of diastereomeric ketals with (*R*,*R*)-butane-2,3-diol, and radio-GC analysis on Superox-20 M. The [^{3}H]-α-pinene from cyclase I was found to be essentially pure (+) enantiomer (≥95%), but contrary to expectation,[6] the product from cyclase II contained equal amounts of (+) and (−) enantiomers. The presence of the (+) enantiomer (presumably produced by a contaminating (+)-pinene cyclase) did not interfere with the radiochemical analysis since it was quantitatively carried through the degradation sequence and quantitatively removed upon crystallization of the final (−)-[^{3}H]diol (**12**).

(17) All intermediates between α-pinene and diol **12** were purified by flash chromatography and/or Kugelrohr distillation. The chemical purity of each was established by GC and TLC analyses and in addition by microanalysis of **8** and **12**.

(18) All reactions in the degradation scheme were optimized by repeated runs with unlabeled compounds. The purity of all intermediates was established by GC, TLC, and, if crystalline, melting point determinations. The structures were confirmed by ^{1}H NMR and IR spectra.

(19) Crandall, J. K.; Crawley, L. C. *Org. Synth.* **1973**, *53*, 17–21.

(20) Boger, D. L.; Mullican, M. D.; Hellberg, M. R.; Patel, M. *J. Org. Chem.* **1985**, *50*, 1904–1911.

(21) The specific activities of all preceding and subsequent intermediates were within ±1–11% of that determined for [^{3}H]-**8**.

(22) Gibson, T. W.; Erman, W. F. *J. Am. Chem. Soc.* **1969**, *91*, 4771–4778.

(23) Firouzabadi, H.; Sardarian, A. R.; Moosavipour, H.; Afshari, G. M. *Synthesis* **1986**, 285–288.

(24) Strictly speaking, these results show only where the tritium label is not located. However, it is very unlikely that any positional scrambling occurred during the enzymatic cyclizations. The principal competing reactions are enzymatic hydrolysis and nonenzymatic solvolysis. Also previous studies[26,8,13] have shown that [1-^{3}H]geranyl pyrophosphate undergoes enzymatic cyclization to [7-^{3}H]pinenes without appreciable positional scrambling.

(25) The radioactivity arising from the presence of the enantiomer[16] has been deducted in order to present values that are directly comparable.

(26) (a) Cane, D. E.; King, G. G. S. *Tetrahedron Lett.* **1976**, 4737–4740. (b) Nozoe, S.; Kobayashi, H.; Morisaki, N. *Ibid.* **1976**, 4625–4626. (c) Corey, E. J.; Cane, D. E.; Libit, L. *J. Am. Chem. Soc.* **1971**, *93*, 7016–7021 and references cited therein.

farnesyl pyrophosphate. However, since the *endo*-bergamotenes[22,27] as well as other sesquiterpenes[28] having the endo stereochemistry are known natural products, the generality of least-motion mechanisms is uncertain at this time.

Acknowledgment. This investigation was supported in part by Grants GM 13956 (R.M.C.) at the University of Illinois and GM 31354 (R.B.C.) at Washington State University from the National Institutes of Health. We thank Michael Satterwhite for the enantiomeric purity determinations and Angela Dillner for assistance with large-scale enzyme isolations.

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(28) (a) Uyehara, T.; Yamada, J.-i.; Kato, T.; Bohlmann, F. *Bull. Chem. Soc. Jpn.* **1985**, *58*, 861–867. (b) Hodgson, G.; MacSweeney, D. F.; Mills, R. W.; Money, T. *J. Chem. Soc., Chem. Commun.* **1973**, 235–236. (c) Christenson, P. A.; Willis, B. J. *J. Org. Chem.* **1979**, *44*, 2012–2018.

Effects of Tunneling on NMR Spectra. The Question of Heavy Atom Tunneling in Norbornyl Cations Reexamined

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Recently Dewar and Merz[1] have suggested that the NMR results concerning norbornyl cation can be explained by invoking heavy atom tunneling, as was previously suggested by Fong.[2] Aside from the serious question of whether the theoretical method employed by them is able to give anything like an adequate prediction of the tunneling frequency, it is relevant to consider what the observable effects of tunneling would be if it *did* occur. Johnson and Mottley[3,4] studied the solid-state proton spectrum in 1,1-dideuterioethyl iodide, where light atom tunneling must be considered in the rotation of the methyl group. At 127 K they observed broadening in the methyl spectrum, which they ascribed to thermally activated passage over the barrier. At 87 K, the spectrum changed to that expected for a fixed methyl group. However, on further cooling to 4.2 K, a number of additional lines appeared which were interpreted as clearly being due to tunneling. A tunneling frequency of 21 kHz was used to calculate a theoretical spectrum which fit the observed spectrum. The apparent tunneling frequency was found to strongly decrease with increasing temperature! It is a common feature of such systems that free quantum tunneling effects are averaged out as the temperature increases and thermally driven incoherent processes dominate at higher temperature.[5]

An important point[6] is that tunneling alone *does not* lead to the familiar broadening phenomena in the NMR but instead produces shifts and new lines. To produce broadening and coalescence, an incoherent, random process such as thermal passage over a barrier is needed. In contrast, tunneling, a coherent process, leads to splitting of states, which yields shifted and new lines, all sharp. As a model for this process, we consider two spin $^1/_2$ nuclei with chemical shifts separated by δ but no dipole–dipole interactions. The spatial motion is constrained by a symmetrical double potential well such that the tunneling splitting of the ground state is ΔE and the rate at which the spin tunnels to the adjacent well is $\nu_T = 2\Delta E/h$. A simple calculation of the NMR spectrum was performed in the spirit of the previous calculation for CH_2D with basis functions of the form $|i\rangle = G(x - x_j)|\sigma\rangle$ where $G(x \simeq x_j)$ is a sharply peaked spatial function centered at x_j ($j = 1, 2$), σ represents the spin state α or β, and $i = 1$–4. The results, details of which are published elsewhere,[7] are illustrated in Figure 1. If norbornyl had a double-minimum surface with tunneling in the appropriate frequency range, such spectra would be expected. If the apparent tunneling frequency decreased at higher temperature, as in the methyl case, and thermal passage over the barrier increased in rate, the side peaks due to tunneling might shift and broaden, and substantial broadening might be expected in the central peak. Since *no* such splitting appeared in the spectrum of norbornyl cation at 4 K observed by Yannoni, Macho, and Myhre,[8] nor was any broadening seen at higher temperatures, it can be concluded that tunneling is not an adequate explanation of these results. The simplest conclusion is that norbornyl has a single-minimum energy surface. Only very high frequency tunneling at all temperatures or an extremely low barrier permitting rapid thermal passage at very low temperature might also be consistent with the spectra. The calculations reported here suggest that NMR might be useful for detecting tunneling effects associated with rearrangements or conformational changes at low temperature; however, intermolecular interactions in solids might well destroy the symmetry of the expected double-well potential curves and thus attenuate the effect. This expected attenuation also makes it less likely that the single line seen in the low-temperature spectrum of norbornyl is due to tunneling.

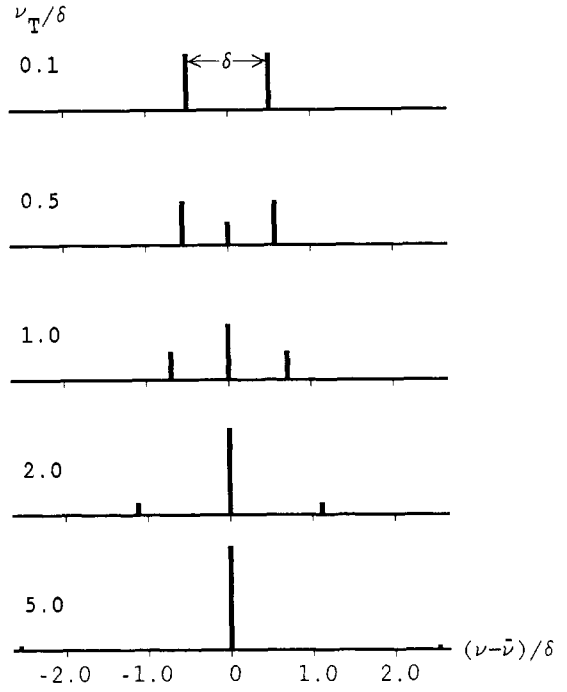

Figure 1. Calculated NMR spectra for a spin $^1/_2$ nucleus in a symmetrical double-well potential for various values of tunneling frequency, ν_T.

The isotopic perturbation results[9] also cannot be explained by tunneling. In the first place, the experiments were done at much higher temperatures where the apparent tunneling frequency in the NMR might be expected to be much smaller even if tunneling

(1) Dewar, M. J. S.; Merz K. M.; *J. Am. Chem. Soc.* **1986**, *108*, 5634.
(2) Fong, F. K.; *J. Am. Chem. Soc.* **1974**, *96*, 7638.
(3) Johnson, C. S., Jr.; Mottley, C. *Chem. Phys. Lett.* **1973**, *22*, 430.
(4) Johnson, C. S., Jr.; Mottley, C. *J. Phys. C.* **1976**, *9*, 2789.
(5) Clough, S.; McDonald, P. J.; Zelaye, F. O. *J. Phys. C* **1984**, *17*, 4413.
(6) Johnson, C. S., Jr. *J. Chem. Phys.* **1973**, *59*, 623.
(7) Johnson, C. S., Jr. *J. Magn. Reson.*, in press.
(8) Yannoni, C. S.; Macho V.; Myhre, P. C. *J. Am. Chem. Soc.* **1982**, *104*, 7380.
(9) Saunders, M.; Kates, M. H.; *J. Am. Chem. Soc.* **1980**, *102*, 6867.

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