(*c* 2, 10% HCl); cmr[9] (H_2O–D_2O, 9:1, neutralized with NaOH) δ 17.6 ppm (methyl) was strongly labeled whereas δ 18.8 ppm (methyl) was unlabeled.

The stereospecificity of our synthesis is supported by the cmr spectrum of the final product, which exhibited only one ^{13}C-labeled methyl signal. The assignment of the 2*S*,3*S* configuration is consistent with the mode of synthesis from (2*S*,3*R*)-3-[^{13}C]methylaspartic acid.[10] The configuration of the β center was correlated with asymmetrically labeled [4-$^2H_3$]valine prepared by Professor R. K. Hill,[11] who observed that the (*R*)-methyl possessed a lower chemical shift than the (*S*)-methyl of valine. When unlabeled **14** was reduced with deuterium gas, the pmr signal of the lower field (*R*)-methyl of the resulting valine was reduced in intensity indicating that the γ-carboxyl group in enzymatic 3-methylaspartic acid corresponds to the (*R*)-methyl of L-valine, thus confirming the 3*S* assignment of the ^{13}C-labeled valine.

(2*S*,3*S*)-[4-^{13}C]Valine (from 100 mg of hydrochloride neutralized with NaOH) was added in eight portions to washed cells of *C. acremonium* (60 g) in shake flasks during 10 hr. The filtrate was freeze-dried and the residue was chromatographed over a Sephadex G-25 (fine grade) column. Elution of the column with isopropyl alcohol–H_2O (7:3) afforded 25 mg of crude penicillin N (79% pure by bioassay). In a similar experiment, 4.1 mg of crude cephalosporin C was obtained.

The cmr spectrum of the cephalosporin C sample showed that the methylene carbon at C-17 (δ 65.0 ppm)[12] was very strongly labeled (signal:noise, 30:1). None of the contaminant signals[13] corresponded to carbons of cephalosporin C. The cmr spectral data of penicillin N are listed in Table I. The chemical-shift assignments of the side-chain carbons of penicillin N were made by a direct correlation with those of cephalosporin C.[12] Other signals were assigned by correlations with published penicillin cmr spectra.[14] The α-methyl carbon appeared to be labeled >20%. No significant amount of label above natural abundance was observed of the β-methyl carbon. The pmr spectrum of the ^{13}C-labeled penicillin N further corroborates this conclusion, for the upfield methyl signal, δ 1.32 ppm (α-methyl by comparison with published data[15]), is reduced in intensity, and the two ^{13}C-satellite signals ($J(^{13}C-^1H)$ = 128.4 Hz) are intense.

These experimental data clearly show that the (2*S*,3*S*)-[4-^{13}C]valine is incorporated into penicillin N with retention of configuration. In the case of cephalosporin C, the ^{13}C label is located in the exocyclic methylene carbon (C-17).[16]

Table I. ^{13}C Chemical-Shift Assignments[a] of Some β-Lactam Antibiotics

Assignment	Chemical shifts: Penicillin N	Methyl-6-acetamido penicillinate[c]	Cephalosporin C: Present work	Lit.[d]
C-11	21.8		35.5	34.7
C-α	27.3	26.1		
C-β	30.9[b]	30.0		
C-12	30.7[b]	30.0	21.8	20.2
C-10	35.4			
C-13	55.4		30.7	29.7
C-6	58.8	58.3		
C-2	65.1	63.6		
C-5	67.4	67.1		
C-3	74.0	69.7		
C-7	175.3[b]			
C-9	175.4[b]			
C-14	175.2[b]		55.5	54.9
C-15	176.7[b]			

[a] Chemical shifts were measured relative to internal *p*-dioxane and corrected to $(CH_3)_4Si$ as internal reference by the relationship $\delta_s(CH_3)_4Si = \delta(p\text{-dioxane}) + 67.4$ (J. B. Stothers, "Carbon-13 NMR Spectroscopy," Academic Press, New York, N. Y., 1972, p 49). [b] The relative assignments have not been made of these carbons. [c] Reference 14. [d] Reference 12.

Acknowledgment. This investigation was supported by research grants from the National Institutes of Health (AI-10,519). The authors are indebted to Professor Richard K. Hill, who kindly informed us in detail of his synthesis and stereochemical assignments of asymmetrically labeled valine-*4*-d_3 in advance of publication, Professor D. Arigoni for valuable suggestions, and Mr. John Usher for a sample of purified unlabeled penicillin N. We acknowledge the support of the Graduate School of the University of Wisconsin for purchase of the nmr spectrometer and pulse Fourier transform accessories.

(9) The carbon magnetic resonance (cmr) spectra were recorded at 22.63 MHz in the Fourier transform mode of operation with proton broad band decoupling on a modified Bruker HX-90E using 10-mm sample tubes. Ten per cent D_2O served as the lock signal and all chemical shifts are given in parts per million relative to tetramethylsilane using *p*-dioxane as the internal standard. The natural abundance cmr spectra of valine were reported by W. Horsley, H. Sternlicht, and J. S. Cohen, *J. Amer. Chem. Soc.*, 92, 680 (1970).

(10) H. A. Barker, R. D. Smyth, E. J. Wawszkiewicz, M. N. Lee, and R. M. Wilson, *Arch. Biochem. Biophys.*, 78, 468 (1958).

(11) Private communication.

(12) A natural abundance cmr spectrum of cephalosporin C was published by N. Neuss, C. H. Nash, P. A. Lemke, and J. B. Grutzer, *J. Amer. Chem. Soc.*, 93, 2337, 5314 (correction) (1971).

(13) The signals of the contaminants were at δ 27.3 (α-CH_3 of penicillin N), 28.6 (penicillin N decomposition product), and 73.3 ppm (unknown).

(14) R. A. Archer, R. D. G. Cooper, and P. V. Demarco, *Chem. Commun.*, 1291 (1970).

(15) See ref 1, pp 686–703.

(16) Since the submission of this manuscript, two communications on this same topic have appeared (see J. E. Baldwin, J. Löliger, W. Rastetter, N. Neuss, L. L. Huckstep, and N. De La Higuera, *J. Amer. Chem. Soc.*, 95, 3796 (1973); N. Neuss, C. H. Nash, J. E. Baldwin, P. A. Lemke, and J. B. Grutzner, *ibid.*, 95, 3797 (1973)). Dr. Neuss has kindly informed us that the correct nomenclature of their synthetic chiral valine is (2*RS*,3*R*)-[4-^{13}C]valine. It is gratifying to note the complementary results of the two incorporations (2*S*,3*R*)-[4-^{13}C]valine (Baldwin) and (2*S*,3*S*)-[4-^{13}C]valine (this paper).

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Stereochemical Dependence of the Chemical-Shift Isotope Effect

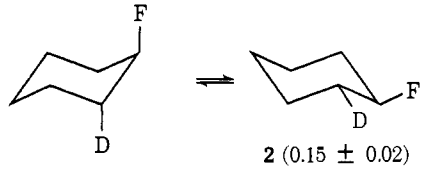
Sir:

Dependence of the chemical shift of a nucleus on the isotopic identity of neighboring nuclei was initially observed in 1953 for the case of the hydrogen molecule.[1] Despite an increasing amount of empirical data, the cause of isotope shifts in polyatomic mole-

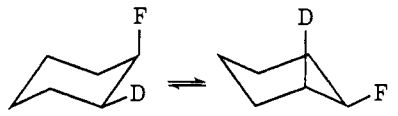
(1) T. F. Wimett, *Phys. Rev.*, 91, 476 (1953).

cules is still poorly understood.[2,3] The isotope shift most frequently encountered is the geminal effect, X–C–Y, in which the shielding of Y depends on the isotopic identity of X, *e.g.*, hydrogen or deuterium. Less thoroughly studied, presumably because of its low magnitude, has been the vicinal isotope effect: X–C–C–Y. In order to test some of the theories forwarded to explain polyatomic chemical-shift isotope effects, we have prepared a series of molecules containing the D–C–C–F fragment with differing known torsional arrangements about the saturated C–C bond. We wish to report that the vicinal isotope effect has a strong stereochemical dependence that parallels the previously known dependence of the β secondary kinetic deuterium isotope effect.[4] The chemical-shift isotope effect is twice as large when the dihedral angle is 180° as when it is 60°. These observations place certain constraints on the theories developed to explain this phenomenon.

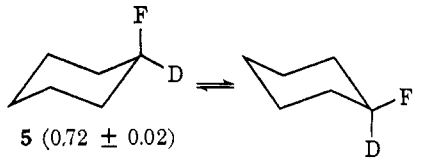
Molecules **1–4** were prepared stereospecifically, and the vicinal isotope effect on the ^{19}F resonance was measured by comparison with the analogous undeuterated compound, cyclohexyl fluoride (**7**, **8**). The



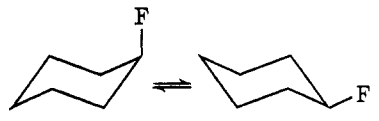
1 (0.35 ± 0.02 ppm) **2** (0.15 ± 0.02)



3 (0.18 ± 0.03) **4** (0.13 ± 0.02)

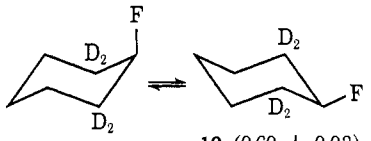


5 (0.72 ± 0.02) **6** (0.81 ± 0.02)



7 (0.0) **8** (0.0)

geminal effect was similarly evaluated with compounds **5** and **6**. The fluorides were prepared from the appropriate cyclohexanols with inversion of configuration by treatment with 2-chloro-1,1,2-trifluorotriethylamine, $(C_2H_5)_2NCF_2CHClF$.[5] The cis alcohol precursor of the trans pair of fluorides, **1** and **2**, was obtained by deuteroboration of cyclohexene. Reduction of cyclohexene oxide with $LiAlD_4$ gave the trans alcohol, precursor of the cis fluorides. Reduction of cyclohexanone with $LiAlD_4$ gave the geminally deuterated alcohol that led to **6** and **7**. To test the additivity of the isotope effect, the cyclohexyl-*2,2,6,6-d*$_4$ fluorides (**9** and **10**)



9 (1.06 ± 0.03) **10** (0.60 ± 0.03)

were prepared from fully exchanged cyclohexanone by way of the alcohol. For measurement of the chemical-shift isotope effect of the individual axial and equatorial isomers, ^{19}F spectra (84.6 MHz) were taken under conditions of slow ring reversal in $CFCl_3$ at −85°, with the ^{19}F chemical shifts referred to internal CF_3CCl_3. The shift in parts per million from the undeuterated material (**7** or **8**) is given under each structure. All shifts are to higher field and are the average of at least five determinations. The primary source of error in the measurement is the large line width of the peaks, since decoupling was not utilized.

The chemical-shift isotope effect arises because of differences between the zero-point vibrational energies of the hydrogen and deuterium systems. Nuclear shielding can be altered either because the lower zero-point energy of deuterium within an anharmonic potential well gives rise to different molecular geometries or because the lower mean-square amplitude of the deuterium vibration even within a harmonic potential well changes the electronic distribution.[2,3] The organic chemist usually discusses these zero-point differences in terms of inductive, steric, hyperconjugative, etc., effects.[6] Changes in the chemical shift associated with substitution of deuterium for a neighboring hydrogen have been variously attributed to variations in electric fields, through bond induction, and hybridization.[3] These explanations are examined below in the light of our data from the cyclohexyl fluorides. It has also been suggested that thermal population of excited vibrational states can be the cause of this isotope effect.[7] The recent evidence of Brey, *et al.*, however, appears to exclude this explanation.[8]

The isotope shift cannot be attributed to a distance-dependent but angular-independent field effect, since our largest vicinal shift (0.35 ppm in **1**) occurs in the compound in which the resonating nucleus and the perturbing isotope are furthest apart. Thus, strictly distance-dependent electric-field or van der Waals effects are excluded. The observed isotope shifts may be attributed to an angular-dependent inductive effect[9] or change in molecular structure (effects that may not be unrelated). The concept of vicinal (σ) delocalization[10] presents a possible explanation for the observed angular dependence.[11] The maximum bond order between vicinal hydrogens in a saturated H–C–C–H fragment was found to occur when the atoms are antiperiplanar,[10] so that substitution of deuterium in this arrangement would exert a maximal effect.

(2) T. W. Marshall, *Mol. Phys.*, **4**, 61 (1961).
(3) (a) H. Batiz-Hernandez and R. A. Bernheim, *Progr. Nucl. Magn. Resonance Spectrosc.*, **3**, 63 (1967); (b) Y. Kanazawa, J. D. Baldeschwieler, and N. C. Craig, *J. Mol. Spectrosc.*, **16**, 325 (1965).
(4) V. J. Shiner, Jr., and J. G. Jewett, *J. Amer. Chem. Soc.*, **86**, 945 (1964).
(5) E. L. Eliel and R. J. L. Martin, *J. Amer. Chem. Soc.*, **90**, 682 (1968).
(6) M. Wolfsberg, *Accounts Chem. Res.*, **5**, 225 (1972).
(7) L. Petrakis and C. H. Sederholm, *J. Chem. Phys.*, **35**, 1174 (1961).
(8) W. S. Brey, Jr., K. H. Ladner, R. E. Block, and W. A. Tallon, *J. Magn. Resonance*, **8**, 406 (1973).
(9) L. Phillips and V. Wray, *J. Chem. Soc., Perkin Trans. 2*, 220, 223 (1972).
(10) J. A. Pople and D. P. Santry, *Mol. Phys.*, **7**, 269 (1963).
(11) Such an explanation has been forwarded in a different context; M. H. Pendlebury and L. Phillips, *Org. Magn. Resonance*, **4**, 529 (1972).

The observed vicinal isotope shifts are additive, within the experimental error. The shift of the axial fluorine in the d_4 compound (**9**, 1.06 ± 0.03 ppm) compares well with the sum of two axial–axial and two axial–equatorial shifts (0.98 ± 0.11). The shift of the equatorial fluorine in **10** (0.60 ± 0.03) is close to the sum of two equatorial–equatorial and two equatorial–axial shifts (0.66 ± 0.07). This near additivity contrasts with observations in a geminal series, CH_4, CDH_3, CD_2H_2, CD_3H, in which the isotope shifts for the last three compounds are 0.019, 0.027, and 0.045 ppm.[12] It is therefore not valid at this point to generalize from our vicinal ^{19}F isotope shifts to other cases, such as these smaller geminal ^1H shifts.

Acknowledgment. This work was supported by the National Science Foundation (Grants GP-34259X and GP-35868X).

(12) R. A. Bernheim and B. J. Lavery, *J. Chem. Phys.*, **42**, 1464 (1965).

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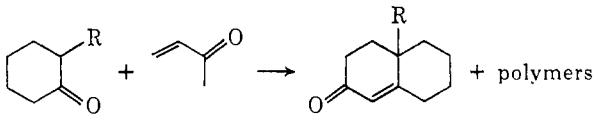
α-Silylated Vinyl Ketones. A New Class of Reagents for the Annelation of Ketones

Sir:

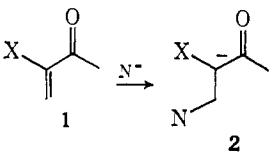
The annelation of 2-alkylcyclohexanones with methyl vinyl ketone and its homologs is an important route to fused polycyclic systems.[1] While condensation at the less substituted α carbon can be carried out efficiently using the corresponding enamines,[2] the conditions required for reaction at the more substituted site result in polymerization of the vinyl ketone and generally low yields.[3] This type of difficulty is avoided



by the use of alkyl halides instead of vinyl ketones[4] but a number of steps are then required to transform the added alkyl group into the desired 3-ketoalkyl function.

The problem with simple vinyl ketones, such as methyl vinyl ketone, comes from the similar base strengths and reactivities of the enolate ions derived from the starting material and the Michael adduct. We have therefore looked into the possibility of designing stable α-substituted vinyl ketones of type **1**, in which the substituent X would be capable of stabilizing the enolate **2** resulting from 1,4 addition. At the same time X must be easily removed, *e.g.*, during the subsequent cyclization of the adduct.

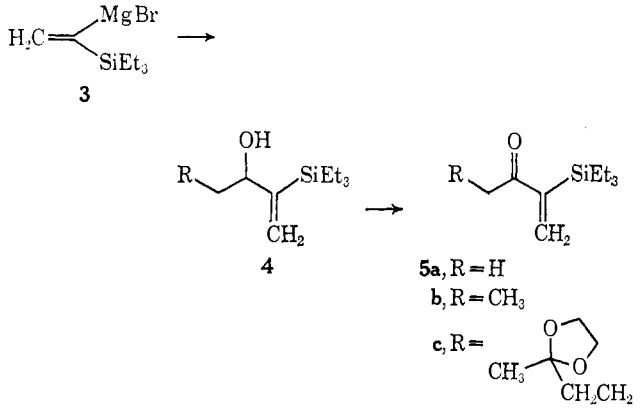
There is good evidence that silicon, with its vacant



1 2

3d orbitals, can stabilize an adjacent negative charge.[5] We wish to report here the synthesis of silylated vinyl ketones such as **1** (X = R_3Si) and illustrate their use in the annelation of certain ketone enolates.

The Grignard reagent **3**, prepared in the usual fashion from α-bromovinyltriethylsilane,[6] reacted with excess acetaldehyde to form **4a** in 80% yield: nmr δ 5.95 (m, 1 H), 5.30 (d, 1 H, J = 2 Hz), 4.35 (br s, 1 H), 1.20 (d, 3 H, J = 7 Hz), 0.4–1.2 (m, 15 H). This allylic alcohol was readily oxidized by Jones reagent to the enone **5a**: bp 83° (7 mm); nmr δ 6.58,



6.11 (AB quartet, J = 2 Hz), 2.15 (s, 3 H), 0.4–1.2 (m, 15 H); ir 5.99 μ; mass spectrum m/e 184 (M^+).[7]

In a similar fashion, the corresponding vinyl ketones **5b** (bp 100° (7 mm)) and **5c** were prepared from propionaldehyde and 5-ethylenedioxyhexanal,[8,9] respectively. Besides being pleasant-smelling liquids, these α-silylated vinyl ketones could be distilled with virtually no decomposition and proved to be stable for months at −20°.

We have made the important observation that α-silylated enones can participate in Michael addition under *aprotic conditions*. These are, of course, the conditions which lead to maximum polymerization with methyl vinyl ketone itself. The lithium enolate of cyclohexanone (generated in tetrahydrofuran from its enol silyl ether, using methyllithium)[10] was treated with 1 equiv of **5a** at −78° and the solution was then allowed to reach room temperature; this led to the adduct **6** in high yield.[11] The validity of the approach

(1) For a good review see E. D. Bergmann, D. Ginsburg, and R. Pappo, *Org. React.*, **10**, 179 (1959).
(2) G. Stork, A. Brizzolara, H. Landesman, J. Szmuszkovicz, and R. Terrell, *J. Amer. Chem. Soc.*, **85**, 207 (1963).
(3) J. A. Marshall and W. I. Fanta, *J. Org. Chem.*, **29**, 2501 (1964), and references cited therein.
(4) G. Stork, S. Danishefsky, and M. Ohashi, *J. Amer. Chem. Soc.*, **89**, 5459 (1967); G. Stork, S. Uyeo, T. Wakamatsu, P. Grieco, and J. Labovitz, *J. Amer. Chem. Soc.*, **93**, 4945 (1971).
(5) Vinyltriarylsilanes, for example, readily add organolithium reagents: (a) L. F. Cason and H. G. Brooks, *J. Amer. Chem. Soc.*, **74**, 4582 (1952); (b) L. F. Cason and H. G. Brooks, *J. Org. Chem.*, **19**, 1278 (1954). Also, tetramethylsilane has recently been metalated: (c) D. J. Peterson, *J. Organometal. Chem.*, **9**, 373 (1967).
(6) Prepared by the method of A. Ottolenghi, M. Fridkin, and A. Zilkha, *Can. J. Chem.*, **41**, 2977 (1963).
(7) This substance was obtained pure only by preparative gas chromatography. The crude distilled product could be used in annelations.
(8) This aldehyde was prepared by reduction of the corresponding nitrile with diisobutylaluminum hydride. The silyl vinyl ketone **5c** was purified by silica gel chromatography using benzene.
(9) Nmr, ir, and mass spectral data are consistent with the proposed structure.
(10) G. Stork and P. F. Hudrlik, *J. Amer. Chem. Soc.*, **90**, 4462, 4464 (1968); H. O. House, L. J. Czuba, M. Gall, and H. D. Olmstead, *J. Org. Chem.*, **34**, 2324 (1969).
(11) By comparison, the reaction of methyl vinyl ketone under identical conditions gave less than 5% of material arising from 1,4 addition.