urations that differ in just the assignment of electrons among the D_{3h} nonbonding orbitals[2] generates only the symmetry-correct D_{3h} wave functions for **2**.[3] Because these ionic terms are absent from the allyl plus p wave function, CI is less important in improving it.

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Conformational Studies of Pantetheine and the Pantetheine Moiety of Coenzyme A

Sir:

In recognition of the essential roles of coenzyme A in intermediary metabolism and numerous other biosynthetic pathways, considerable effort has been expended to establish the specificities of enzymes for various portions of the molecular unit.[1] Partially because no crystalline derivative of CoA has been investigated by diffraction techniques, there has been little speculation and no published work relating the conformation of CoA or its analogs to binding and specificity in enzyme systems requiring it as a cofactor. We now report our initial results in the area of conformational analysis, which help to define aspects of the preferred conformation of coenzyme A in aqueous solution based upon coupling constants derived from NMR spectra of CoA (Figure 1) and pantetheine (Figure 2).

Vicinal coupling in isolated, rapidly rotating ethylene units such as those of the β-alanine and cysteamine portions of coenzyme A is characterized by two coupling constants (J and J') commonly defined[2] by

$$J = n_t J_g + \tfrac{1}{2} n_g (J_t + J_g)$$

$$J' = n_t J_t + n_g J_g$$

where n_t and n_g are the mole fractions of the classical staggered rotamers in which the coupled vicinal atoms are trans and gauche, respectively, and where J_g and J_t are characteristic vicinal coupling constants associated with dihedral angles near 60 and 180°. Rotamer populations of isolated ethylene groups for which deceptively simple triplet AA'XX' patterns are often observed may be obtained by applying the condition $n_t + n_g = 1$ to the previous equations to provide

$$n_t = 2[(J + J') - \tfrac{1}{2}(J_t + 3J_g)]/(J_t - J_g)$$

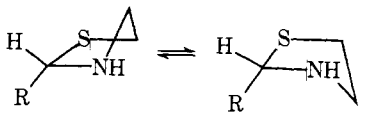
Thus the mole fraction of the trans rotamers of the cysteamine and β-alanine moieties may be obtained from the average coupling constants, $(J + J')/2$, measured for the triplet patterns of CoA and its derivatives at 220 MHz (Table I) provided the two additional coupling constants, J_g and J_t, are available for each moiety.

Table I. Coupling Constants and Mole Fraction of Trans Rotamer in the Cysteamine and β-Alanyl Moieties of CoA and Pantetheine

Compound	Temp, °C	Solvent	β-Alanyl (Hz) J_{av}	β-Alanyl (Hz) n_t	Cysteamine (Hz) J_{av}	Cysteamine (Hz) n_t
Pantetheine	18	D_2O	6.40	0.34	6.50	0.42
	62	D_2O	6.50	0.42	6.55	0.46
	18	DMSO-d_6	6.9	0.74	*a*	
Coenzyme A	18	D_2O	6.13	0.19	6.72	0.60
	63	D_2O	6.68	0.58	6.62	0.55
	18	DMSO-d_6	6.9	0.74	*a*	

[a] Interference by DMSO resonance.

For the cysteamine moiety the value of $J_g = 4.71$ Hz was obtained from the spectrum of 1,3-thiazolidine[3] in which by analogy to the 1,3-oxathiolanes[4] only two equally populated pseudo-rotamers Ia and IIa with staggered ethylene units



Ia, R = H
Ib, R = *t*-Bu

IIa

are expected to be populated. The value for $J_t = 9.77$ Hz was taken from the largest coupling constant of the ABCD spectrum of 2-*tert*-butyl-1,3-thiazolidine for which only a single pseudorotamer Ib is expected to be significantly populated.

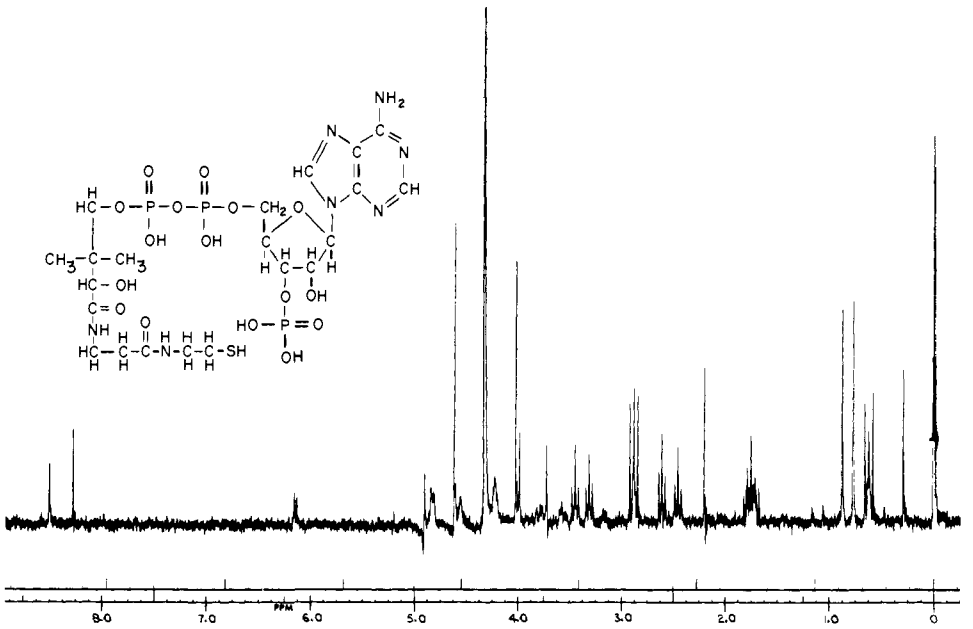


Figure 1. The 220-MHz NMR spectrum of coenzyme A obtained in D_2O at 63°. Methylene resonances of the cysteamine portion of the molecule give rise to deformed triplet resonances at 2.58 (CH_2S) and 3.29 ppm downfield from internal DSS. Deformed triplets occur at 2.43 (CH_2CO) and 3.43 ppm for the β-alanine moiety. The average coupling constants were measured on expanded spectra.

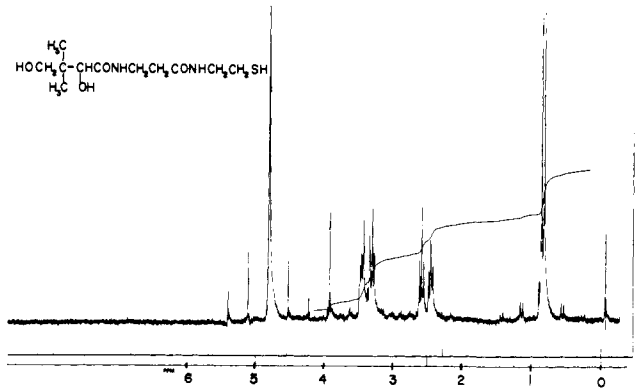


Figure 2. The 220-MHz NMR spectrum of pantetheine in D_2O solution at 18°. Methylene resonances of the cysteamine portion of the molecule are seen as deformed triplets at 2.65 (CH_2S) and 3.30 ppm downfield from internal TSP. Deformed triplets occur at 2.51 (CH_2CO) and 3.45 ppm for the β-alanine residue. The average coupling constants were measured on expanded spectra.

Table II. Enthalpy and Entropy Changes for the Rotameric Equilibrium Gauche ⇋ Trans for CoA and Pantetheine

		β-Alanyl moiety		Cysteamine moiety	
Compound	Solvent	ΔH, kcal/mol	ΔS, eu	ΔH, kcal/mol	ΔS, eu
CoA[a]	D_2O	7.6 ± 2.2	23	−0.88 ± 0.26	−2
Pantetheine[a]	D_2O	1.44 ± 0.4	25	0.71 ± 0.03	2
β-Mercapto-ethylamine[b]	$DCCl_3$			−0.49 ± 0.24	
β-Alanine[b]	$DCCl_3$	−0.43 ± 0.15			

[a] Calculated from rotamer populations at two temperatures, 63 and 18°, using $\Delta G = RT \ln K_{eq} = \Delta H - T\Delta S$ where $K_{eq} = n_t/n_g$. [b] Calculated from observed rotamer populations at 34°, assuming $\Delta S = R \ln 2$.

For the β-alanine moiety we have adopted the values of J_g and J_t used for the cysteamine moiety. This seemed reasonable based upon the known dependence of the magnitude of the average vicinal coupling constant on the sum of the atom electronegativities about the carbons bearing the coupled protons;[5] the electronegativities of carbon and sulfur are identical. Indeed, the adopted values accurately predicted J_{av} for the gauche $NCH_2CH_2C{=}O$ fragment of *N-tert*-butyl-4-piperidone $((J + J')/2)_{calcd} = 6.0$; obsd = 6.0 Hz.

Using the above values for J_g and J_t the rotamer populations (Table I), and entropy and enthalpy changes (Table II) associated with the rotameric equilibria in CoA and pantetheine were calculated. For the β-alanyl moiety of CoA the data provide strong evidence for a highly favored gauche orientation, counter to that reported for β-alanine.[6] Also for the β-alanyl moiety of pantetheine at room temperature, its sensitivity to temperature changes further indicates an enthalpy favoring the gauche rotamer. The magnitudes of ΔH in both CoA and pantetheine, large for ethylene fragments with no attached bulky groups, is consistent with observed enthalpy differences arising from hydrogen bonding. For the cysteamine portion of both CoA and pantetheine the data in Table I show a rotational preference for a trans orientation. Here the enthalpy difference between the two states is about the same as that for cysteamine, consistent with normal rotational preferences and inconsistent with intramolecular hydrogen bonding as a conformation-determining factor for this portion of the molecule. Also consistent with a hydrogen bonding hypothesis is the fact that in DMSO-d_6, a strong disrupter of hydrogen bonds, the trans rotamer is strongly preferred in both pantetheine and CoA for the β-alanine moiety.

Although conformations involving hydrogen bonding between the β-alanyl N–H hydrogen atom and a phosphate oxygen atom base, or sugar hydroxyl groups must be considered for CoA because of the much larger ΔH for the β-alanyl rotamer equilibrium as compared to that of pantetheine, the evidence for a preferred gauche conformation of the β-alanyl fragment of pantetheine where none of these groups are present strongly suggests that any hydrogen bonding occurs internally within the pantetheine moiety in aqueous solution. It is clear that folded conformations in aqueous medium, maintained primarily by hydrogen-bonding forces proposed here, may be important in the recognition of coenzyme A by enzymes as well as in defining the relative positions of groups at the active sites in known multienzyme complexes utilizing the pantetheine prosthetic group.

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On Purported Trigonal Bipyramidal $Cr(CO)_5$

Sir:

For some years now, the vexed question of the equilibrium geometry of the pentacarbonyls $M(CO)_5$ (M = Cr, Mo, W) would appear to have been solved. Square pyramidal species have been repeatedly described[1-3] and bands formerly attributed to a D_{3h} isomer of $Mo(CO)_5$[1] have been reassigned to polymeric species.[2,3] Very recently, Ozin[4] has presented evidence for a new more stable isomer of $Cr(CO)_5$, formed by vapor deposition matrix isolation methods followed by annealing. It is further claimed that the species described by Turner as $Cr(CO)_5$ in CO and other matrices should be reformulated as $Cr(CO)_5S$, where S is an O-bonded or sideways bonded isocarbonyl, that the more stable isomer of $Cr(CO)_5$ is of trigonal bipyramidal geometry, and that isotopic substitution data confirm this assignment.