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Carbon-13 Nuclear Magnetic Resonance Spectra of the Vitamin B-6 Group

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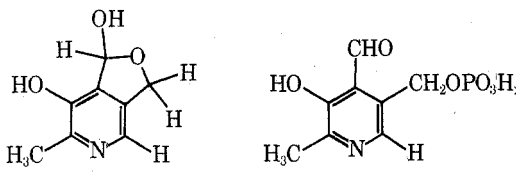
Received December 10, 1974

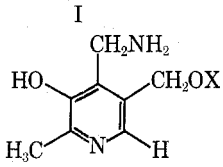
The Fourier transform natural abundance carbon-13 spectra of pyridoxal, pyridoxal 5′-phosphate, pyridoxamine, pyridoxamine 5′-phosphate, and pyridoxine are reported. Resonances are assigned by chemical shift analogies and by spin–spin coupling to adjacent protons. Chemical shifts are strongly pH dependent owing to the deprotonation of the various functional groups. Chemical shift analogies are interpreted as indicating a zwitterionic structure at neutrality. The detailed pH dependence of the carbon-13 chemical shifts of pyridoxal 5′-phosphate, pyridoxamine 5′-phosphate, and pyridoxine are reported. Long- and short-range proton-to-carbon coupling constants are also reported and are not found to be strongly pH dependent.

The vitamin B-6 group is comprised of pyridoxal (PL, I), pyridoxal 5′-phosphate (PLP, II), pyridoxamine (PM, III), pyridoxamine 5′-phosphate (PMP, IV), and pyridoxine (PN, V). These substituted pyridines are essential cofactors to a large number of enzymes involved in the metabolism of amino acids. The reactions they catalyze proceed through Schiff base formation of the amino acid with the substituted 4-pyridine aldehyde form of the vitamin. The Schiff base may undergo any one of a number of electron shifts transforming the amino acid moiety into the various observed products.[1] A particular type of enzyme apparently participates in the reaction by selectively catalyzing only the desired electron shift and by inhibiting all the other types of shifts. In order to understand the means by which enzymes can control the electron shifts in these Schiff bases, it is necessary to understand the factors which influence electron densities in the aromatic ring of the vitamin B-6 Schiff bases. The sensitivity of carbon-13 nuclear magnetic resonance (^{13}C NMR) to electronic structure is well known. We are in the process of carrying out a complete study of the ^{13}C NMR spectroscopy of the vitamin B-6 Schiff bases and their metal complexes, but, as a prelude, it has been necessary to study the vitamin B-6 group itself.

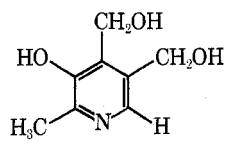


I

II



III, X = H
IV, X = PO_3H_2



V

Herein we report the ^{13}C NMR spectral assignments, pH dependence, and coupling constants for the vitamin B-6 group and some related pyridine derivatives.

Experimental Section

Pyridoxal hydrochloride and pyridoxal-5′-phosphate were purchased from Sigma Chemical Co., and pyridoxamine dihydrochloride was obtained from Mann Research Laboratories. Picoline was from Reilly Tar and Chemical Corp. Pyridoxine, pyridoxamine 5′-phosphate, and the remaining monosubstituted pyridines were from Aldrich. Deuterium oxide was 99.8% from Thompson-Packard; NaOD (40% in D_2O) and DCl (20% in D_2O) were from Diaprep Inc.

Carbon-13 nuclear magnetic resonance spectra were obtained at 40° in Fourier transform mode with a Jeol PS/FFT-100 spectrometer and Jeol EC-100 data system. Chemical shifts were recorded in parts per million relative to an external capillary of dioxane; these values were adjusted to the tetramethylsilane (Me_4Si) scale by adding 67.4 ppm to the observed shift. This ignores a small (<0.2 ppm) correction due to diamagnetic susceptibility.[2] Typically a 6250-KHz range and 16K words of memory were used, giving a digital resolution of 0.76 Hz. The D_2O solvent was the source of an internal deuterium lock. Broad band decoupled (2 KHz), single frequency decoupled, and undecoupled spectra were recorded.

Solutions (1 *M*) of pyridoxal-related compounds (except pyridoxine) in D_2O were maintained at constant ionic strength with sodium chloride (3 *M*); in cases where 1 *M* solutions could not be prepared, saturated solutions were used. The pD (negative logarithm of deuterium ion activity) was recorded by adding 0.41 to the reading of a Brinkmann Model E512 pH meter standardized against aqueous buffers;[3] this method is valid only in the range $2 < pD < 9$, and values outside this range are approximate. No correction was made for sodium ion effects. Acidity was varied by the addition of DCl or NaOD. At these conditions of high salt and reagent concentration, observed pH becomes an ill-defined concept and reported pD values must be viewed as approximate measures of pH.

Results and Discussion

Resonance Assignments. Despite the structural similarity of compounds I–V, complete assignment of their resonances is an arduous task because many fall in a narrow

Table I
^{13}C Chemical Shifts of Pyridoxal and Related Compounds[a]

Compd	pD	CH_3	C-4'	C-5'	C-2	C-3	C-4	C-5	C-6
Pyridoxal (I)	2	15.8	99.6	71.2	144.9	149.9	140.8	139.2	127.0
Pyridoxal 5'-	6.8	17.1	196.9	62.8	152.5	165.6	126.7	137.2	123.4
phosphate (II)	8.7	18.3	197.2	63.1	154.4	166.7	125.9	135.8	125.5
Pyridoxamine (III)	2	16.9	36.1	60.0	144.1	154.3	137.3	139.4	132.6
	11	20.5	37.4	61.2	150.6	161.0	133.6	137.4	133.4
Pyridoxamine 5'-	7.1	16.6	37.6	63.0	145.6	163.6	133.6	135.6	124.7
phosphate (IV)	>11	20.7	37.5	63.9	150.9	161.0	138.2	131.8	134.2
Pyridoxine (V)	2.7	15.4	58.0	59.1	143.6	153.6	141.1	137.7	130.7
	9.9	18.6	58.0	60.3	148.8	161.2	135.5	134.1	130.6

[a] Shifts are in parts per million relative to tetramethylsilane (Me_4Si).

Table II
^{13}C Chemical Shifts for Some Substituted Pyridines in D_2O[a]

Substituent	Degree of protonation	C-2	C-3	C-4	C-5	C-6	Ref	Registry no.
H	0	150.4	124.1	136.1	124.1	150.4	*b*	
	100%	142.6	129.1	148.5	129.1	142.6	*b*	
2-CH_3	0	(159.9)	123.4	137.2	122.0	149.8	*c*	109-06-8
	0	(158.4)	124.7	138.4	122.1	148.4	*d*	
	50%	(156.7)	126.7	142.7	123.6	145.1	*d*	
	100%	(154.2)	129.0	147.6	125.3	141.2	*d*	
3-OH[e]	0	137.8	(153.5)	121.4	123.8	140.0	*f*	109-00-2
	0		(159.9)				*d*	
	100%		(157.4)				*d*	
4-CHO	0	151.5	122.8	(141.9)	122.8	151.5	*g*	872-85-5
	0	151.1	123.6	(142.6)	123.6	151.1	*d*	
4-$CH(OD)_2$	pD 1	142.5	125.6	(162.2)	125.6	142.5	*d*	55298-75-4
4-CH_2OH	pD 10	149.6	122.7	(152.0)	122.7	149.6	*d*	586-95-8
	pD 1	141.8	125.2	(164.3)	125.2	141.8	*d*	
3-CH_2OH[h]	0	148.4	(137.2)	137.4	125.2	148.8	*d*	100-55-0
	pD 5.9	144.9	(139.5)	141.1	126.6	145.6	*d*	
	pD 4.7	140.8	(142.0)	145.6	128.1	141.7	*d*	
	pD 1	140.1	(142.5)	146.3	128.4	141.0	*d*	

[a] δ_C, parts per million from Me_4Si. See ref 2. Data in parentheses are for the substituted carbon. [b] From R. J. Pugmire and D. M. Grant, *J. Am. Chem. Soc.*, **90**, 697 (1968). [c] From P. C. Lauterbur, *J. Chem. Phys.*, **43**, 360 (1968). [d] This work. [e] Only the assignment of C-3 could be made with certainty in aqueous solution because the resonances appeared in a narrow region and shifted with pH. [f] From ref 6. [g] From ref 11a. [h] The assignments of C-2 and C-6 are uncertain.

region of the spectrum and are substantially pD dependent. The following reasoning was used to arrive at the assignments in Table I. In all cases, a single resonance in the region of 15–21 ppm is clearly assignable to the 2'-methyl carbon atom. For pyridoxal and pyridoxal phosphate the remaining carbon atoms which are substituents of the aromatic ring are readily assignable by the multiplicity of their one-bond proton couplings. For pyridoxamine and pyridoxamine phosphate electronegativity arguments suggest that the upfield methylene resonance belongs to the 4' carbon atom while the downfield resonance belongs to the 5' carbon atom. For pyridoxamine phosphate this assignment is verified by the observation of $^2J_{CP} = 3$ Hz to the phosphate group. In all compounds the 5' carbon resonance shows $^3J_{CH} = 3$ Hz via coupling to 6-H. In pyridoxine this coupling was used to assign the resonance at 59–60 ppm to the 5' carbon atom, leaving the resonance at 58 ppm assigned to the 4' carbon atom.

The resonances of the pyrdine ring carbon atoms fall in a fairly narrow range. Here only the assignment of C-6 is straightforward by virtue of its large one-bond coupling to 6-H. In DMSO the 3-carbon atom of 3-hydroxypyridine is found at much lower field than the other carbon resonances.[4] We verified this in D_2O and studied the dependence of chemical shift on pH during deprotonation of the 3-OH group (Table II). The similar chemical shift of the lowest field resonance of all the vitamin B-6 group leads to its assignment as the 3-carbon atom.

Extensive use was made of long-range coupling constants and of single frequency decoupling experiments to arrive at the remaining assignments. In pyridoxal phosphate and pyridoxamine phosphate the 5-C resonance is readily assigned by the observation of three-bond carbon–phosphorus couplings[5] in proton-decoupled spectra. For pyridoxal phosphate, C-4 is also distinctive by virtue of its large (27 Hz) coupling to the aldehyde proton.[6] The remaining unassigned aromatic resonance in pyridoxal phosphate is then C-2 and this must be the resonance at 152–154 ppm which appears as a doublet of quartets via its two-bond coupling to the methyl protons ($J = 4$ Hz) and its three-bond coupling to 6-H ($J = 7$ Hz). For all the vitamin B-6 compounds a resonance of similar structure was observed in a similar region and therefore could be assigned confidently to C-2. These arguments leave all resonances assigned except C-4 and C-5 in the nonphosphorylated compounds. In these compounds, the two resonances are observed within 5 ppm of one another and are observed to cross over upon varying pH in some cases. Assignments of the resonances are difficult and are based on the general observation that two-bond proton–carbon-13 coupling constants are in the range of 1–4 Hz while three-bond couplings are somewhat larger, usually 4–7 Hz.[7] For pyridoxal, the decoupler was set at a

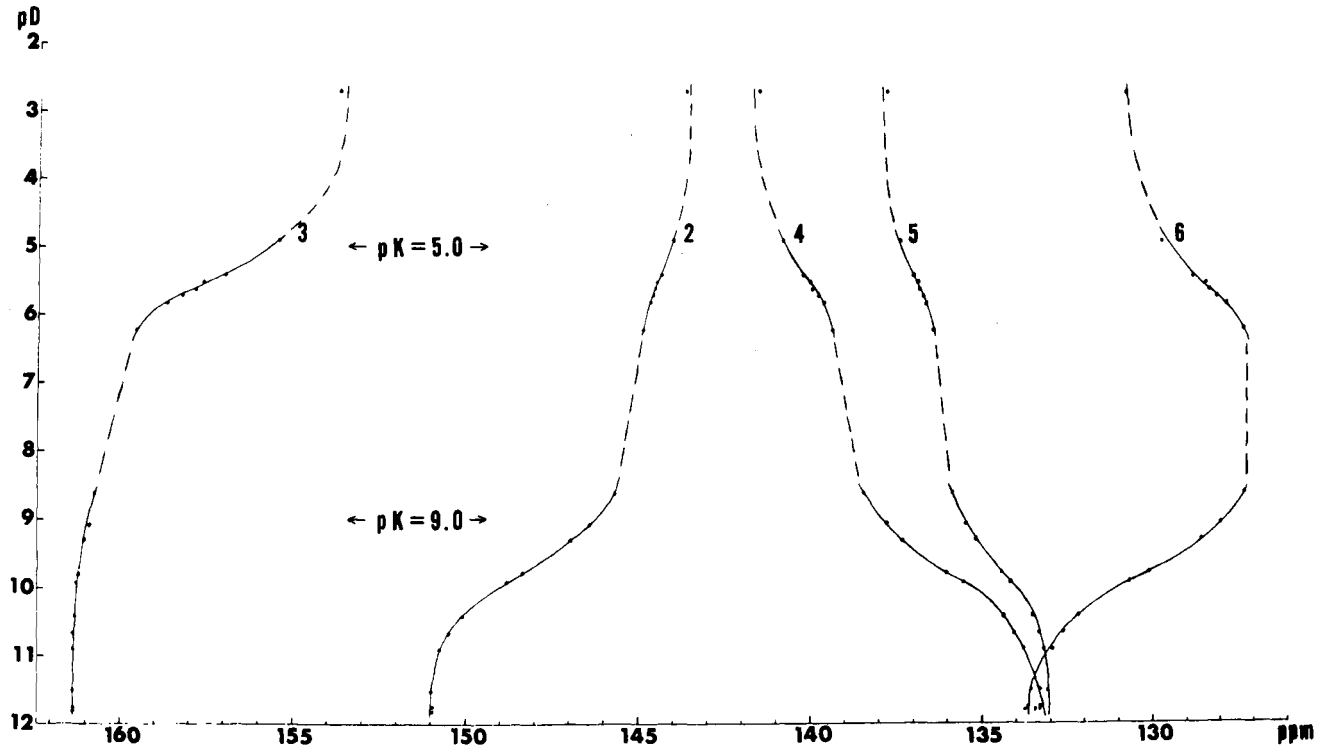


Figure 1. The pD dependence of chemical shifts for the carbon-13 resonances of pyridoxine. The reference is Me_4Si. Arrows refer to reported pK's, determined in H_2O.

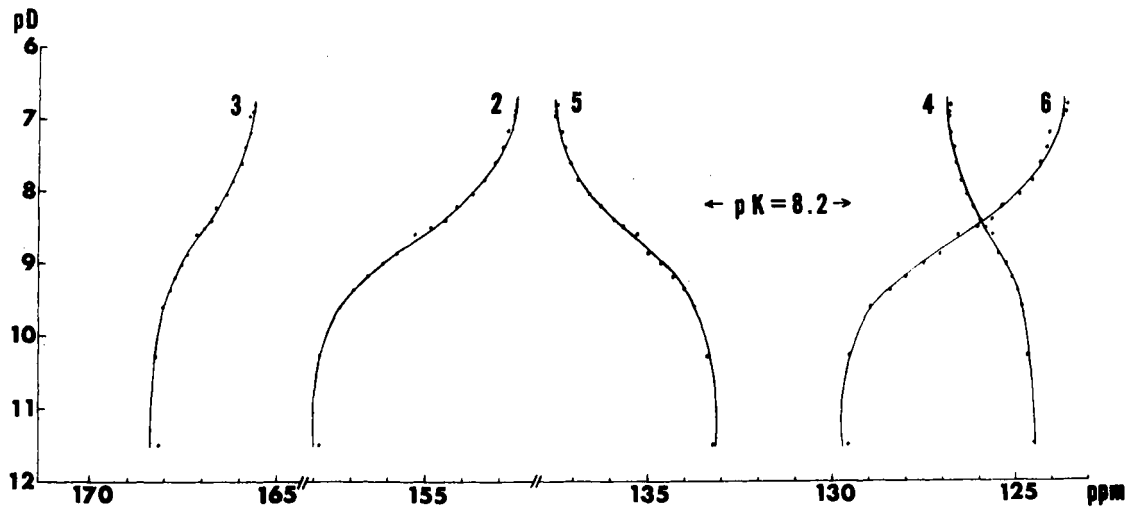


Figure 2. The pD dependence of chemical shifts for the carbon-13 resonances of pyridoxal phosphate. The reference is Me_4Si. Arrows refer to reported pK's, determined in H_2O.

frequency between those of 4′-CH and 5′-CH_2 in the proton spectrum. The decoupling power was adjusted so that 6-C was observed to be nearly completely coupled to 6-H but 4′-C and 5′-C were entirely decoupled from their protons. Under these conditions the resonance at 140.8 ppm is a doublet with J = 5.8 Hz and assigned to 4-C while the resonance at 139.2 ppm is a doublet split by about 2 Hz and is assigned to 5-C. On moving the decoupling frequency toward the 5′-CH_2 resonance, small additional splittings are observed at 4-C while larger ones develop at 5-C. This is indicative of the larger coupling of 5-C to 4′-CH as it becomes inefficiently decoupled. In pyridoxamine, a similar experiment reveals that 4-C and 5-C are reversed and that the resonance at 137.3 ppm belongs to 4-C while that at 139.4 belongs to 5-C. In pyridoxine, the resonances are in the same order as for pyridoxal with 4-C at 141.1 ppm and 5-C at 137.7 Hz.

pH Dependence of Chemical Shifts. Ionization of the pyridinium nitrogen and of the several substituents on the vitamin B-6 ring occurs as the pH of an acidic solution is increased. As expected, this results in a pH dependence of chemical shifts. For the aliphalic carbon nuclei chemical shifts on deprotonation are small—of the order of 1 ppm. Shifts are observed in both upfield and downfield directions with, for example, the 2′-carbon resonance of pyridoxine shifting downfield by 1 ppm while the 4′-carbon resonance shifts upfield by the same amount between pD 2 and pD 9. These and other data for substituent atoms appear in Table I.

The carbon resonances of the pyridine rings are much more profoundly influenced by pH change. Here shifts are as much as 10 ppm over a deprotonation, they occur in either the upfield or downfield directions, and some resonances first shift upfield and then downfield. The data are reported in Figures 1–3. The small differences between the inflection points of these NMR titration curves and the re-

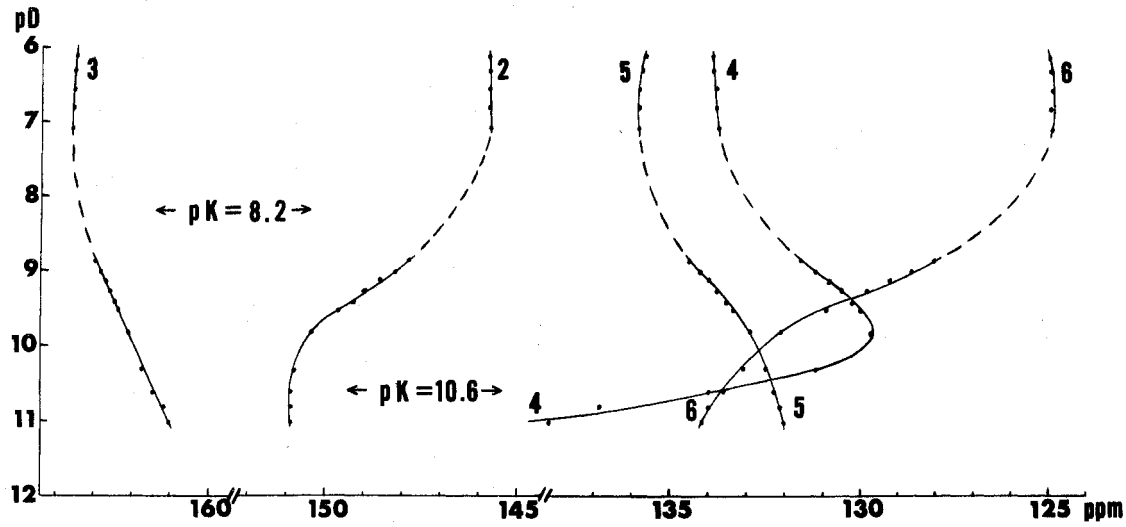
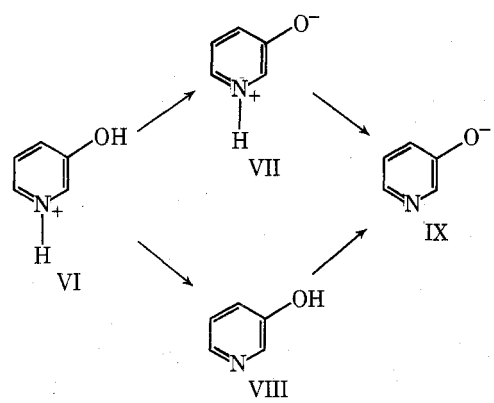


Figure 3. The pD dependence of chemical shifts for the carbon-13 resonances of pyridoxamine phosphate. The reference is Me_4Si. Arrows refer to reported pK's, determined in H_2O for pyridoxamine.

Chart I



ported pK's are due to the fact the pK's were determined at much lower ionic strength and in H_2O rather than D_2O.[8]

For 3-hydroxypyridine derivatives two general pathways for deprotonation are possible. They are represented as VI–IX in Chart I. In neutral solution either the zwitterionic species VII or the neutral species VIII is anticipated. In polar media such as water, VII predominates, although significant amounts of VIII are in equilibrium with it.[9] Therefore, the first deprotonation (pK_a = 4) of the vitamin B-6 group largely corresponds to the deprotonation of the phenolic group while the second corresponds to the deprotonation of the pyridinium nitrogen atom.

In pyridine, deprotonation results in downfield shifts of 2-C and upfield shifts for 3-C and 4-C. The theoretical interpretation for this has been discussed from several points of view.[10] Table II shows that pyridines with individual substituents similar to those of the vitamin B-6 group behave virtually identically to pyridine itself. Figures 1–3 illustrate that fairly similar trends are observed for all but C-3 of the vitamin B-6 group. This resonance shifts to higher field in the high pH deprotonation of pyridoxamine phosphate, to low field with pyridoxal phosphate, and is virtually unchanged for pyridoxine. A possible explanation would be that the different behaviors of 3-C reflect greater or lesser mixing of the 3-OH microscopic deprotonation into the high pK macroscopic deprotonation.

The directions and magnitudes of the pH dependence of chemical shift reflect such important physical effects as changes of electron density and changes in the difference between the energies of the ground and excited states. Since an important way that the enzyme can influence the reactivity of the coenzyme is through selective protonation or deprotonation of the various functional groups, it is particularly important to understand these effects in the vitamin B-6 group. Unfortunately, a detailed rationalization requires an extensive theoretical treatment of the electronic structure and is further complicated by the large and variable solvation energies involved. At the present time, it will suffice to point out that Figure 1 shows that the effect of two deprotonations on 2-C, 4-C, and 6-C is to make their pH dependences distinctive. In the low pH deprotonation 6-C and 4-C shift upfield while 2-C shifts downfield. In the second deprotonation 6-C and 2-C shift downfield while 4-C shifts upfield. Since all three sites are either ortho or para to the deprotonation, such behavior is not explicable by simple arguments; however, it shows that the effect of the substituents is to make the three sites each very different at different pH's. Selective protonation of the two sites can be expected to optimize these differences.

Beyond the effects of deprotonation of the phenolic and pyridinium sites, several other interesting effects are noted. The largest pH dependence of chemical shift is that of the pyridoxamine phosphate's 4-C near the pK for deprotonation of the 4′-$CH_2NH_3^+$. Interestingly, the effects of this deprotonation show up only at this carbon atom and all other adjacent atom are virtually unaffected by it. Also, in the phosphorylated compounds, deprotonations occuring at the phosphate group have almost no influence on the chemical shift of any aromatic carbon atoms in the compounds. Apparently the phosphate deprotonation produces minimal electronic changes in the aromatic system, in contrast to the ammonium deprotonation.

A recent ^{13}C NMR study of 3-hydroxypyridine and related compounds showed great differences between the spectra of the O-methylated and N-methylated isomers.[4] As these correspond to the neutral (VIII) and zwitterionic (VII) forms of the vitamin B-6 group in neutral solution, it is of interest to compare the shifts of these compounds with our data. Table III compares observed shifts for pyridoxal phosphate and for pyridoxine with calculated values predicted from the two methylated 3-hydroxypyridines, using substituent effects observed with monosubstituted pyridines.[11] These substituent shifts were taken from the literature and from the data in Table II. Shifts relative to pyridine were tabulated for each carbon nucleus of the appropriate monosubstituted pyridine. These were added together for all the substituents. Values obtained were added

Table III
Comparison of Observed Carbon-13 Shifts with Values Calculated from Neutral and Zwitterionic Models

	2-C	3-C	4-C	5-C	6-C
Pyridoxal Phosphate					
Observed, pD 6.9	151.5	165.6	126.7	137.2	124.0
Calculated from the neutral model[a]	144.9	152.1	124.8	134.3	141.2
Calculated from the zwitterionic model[b]	142.0	165.7	136.5	136.8	121.2
Pyridoxine					
Observed, pD 6.2	144.8	159.5	139.3	136.3	127.3
Calculated from the neutral model[a]	143.2	152.3	135.7	133.8	139.5
Calculated from the zwitterionic model[b]	140.3	165.8	146.7	136.8	119.5

[a] Calculated by adding substituent effects for monosubstituted pyridines to the chemical shifts of 3-OCH_3 pyridine.[4,b] Calculated by adding substituent effects for monosubstituted pyridines to the chemical shifts of the zwitterion derived by deprotonation of 1-methyl-3-hydroxypyridinium ion.

Table IV
One-Bond Coupling Constants (J_{C-H}) for Pyridoxal and Related Compounds[a]

Compd	pD	C-2'	C-4'	C-5'	C-6
PL	2	131.7	177.8	152.2	194.6
PLP	6.8	129.7	184.6	149.1	193
	8.3	128.2	181.6	148.0	186.9
PM	2	131.5	146.5	146.5	191.5
	11	126.1	136.6	144.6	177
PMP	7.1	130.3	145.0	145.7	188
	>11	126.6	136.2	145.0	177.4
PN	2.7	131.2	146.8	145.7	190.0
	9.9	127.7	144.2	144.2	180.0

[a] In hertz; signs were not determined.

to the reported chemical shifts for the zwitterion resulting from deprotonation of 1-methyl-3-hydroxypyridinium ion and to shifts observed for 3-methoxypyridine, respectively. The CH_2OH group was used as a model for the CH_2OX group.

As discussed above, the vitamin B-6 derivatives are thought to exist in the N-protonated zwitterionic form in neutral solution. The chemical shifts in Table IV generally support this assertion. Observed values are found to be generally quite close to those predicted from the simple model. Some of the discrepancies may be attributed to substantial amounts of the neutral tautomers in equilibrium with the zwitterion, particularly in the case of pyridoxine, where the equilibrium is more important.[9] However, the generally poor agreement for 2-C and 4-C must be attributed to the nonadditivity of substituent effects for these sites.

Coupling Constants. One-bond coupling constants for compounds I–V are listed in Table IV. These coupling constants are controlled by hybridization of the carbon atom. All values of these constants are within reported ranges of related compounds except those of the phosphorylated derivatives. Observed one-bond coupling for methylene carbon atoms uniformly are about 25 Hz less than those reported for the methylene groups of organic phosphates. Evidently, this reflects the interaction of the aromatic ring

Table V
Selected Two- and Three-Bond Coupling Constants for Pyridoxal and Related Compounds[a]

Compd	pD	J^b	
Pyridoxal (I)	2	$^2J_{CCH}$ (methyl to C-2)	6.1
		$^3J_{CNCH}$ (6-H to C-2)	6.1
		$^2J_{CCH}$ (6-H to C-5)	2.3
		$^3J_{CCCH}$ (6-H to C-4)	5.8
Pyridoxal 5'-phosphate (II)	6.8	$^2J_{CCH}$ (methyl to C-2)	6.9
		$^3J_{CNCH}$ (6-H to C-2)	6.9
		$^3J_{CCH}$ (6-H to C-4)	3.6
		$^2J_{CCH}$ (CHO to C-4)	19.2
	8.7	$^2J_{CCH}$ (methyl to C-2)	6.1
		$^3J_{CCCH}$ (6-H to C-4)	6.1
		$^2J_{CCH}$ (CHO to C-4)	21.4
Pyridoxamine (III)	2	$^2J_{CCH}$ (methyl to C-2)	6.5
		$^3J_{CNCH}$ (6-H to C-2)	6.5
		$^3J_{CCCH}$ (5'-methylene to C-6)	4.6
		$^2J_{CCH}$ (6-H to C-5)	2.9
		$^3J_{CCCH}$ (6-H to C-4)	5.0
	11	$^2J_{CCH}$ (methyl to C-2)	6.1
		$^3J_{CNCH}$ (6-H to C-2)	9.9
		$^3J_{CCCH}$ (5'-methylene to C-6)	5.3
Pyridoxamine 5'-phosphate (IV)	7.1	$^2J_{CCH}$ (methyl to C-2)	6.9
		$^3J_{CNCH}$ (6-H to C-2)	6.9
		$^3J_{CCCH}$ (5'-methylene to C-6)	4.6
	>11	$^2J_{CCH}$ (methyl to C-2)	6.1
		$^3J_{CNCH}$ (6-H to C-2)	9.9
		$^3J_{CCCH}$ (5'-methylene to C-6)	4.6
Pyridoxine (V)	2.7	$^2J_{CCH}$ (methyl to C-2)	6.1
		$^3J_{CNCH}$ (6-H to C-2)	6.1
		$^3J_{CCCH}$ (5'-methylene to C-6)	4.2
		$^2J_{CCH}$ (6-H to C-5)	4.9
	9.9	$^2J_{CCH}$ (methyl to C-2)	6.1
		$^3J_{CNCH}$ (6-H to C-2)	9.2
		$^3J_{CCCH}$ (5'-methylene to C-6)	5.3

[a] ±0.8 Hz; signs were not determined.

with the phosphorylated methylene group. In all cases coupling constants decreased upon deprotonation of the compound; however, the changes are not sufficiently great to indicate a strikingly different electronic structure for the pyridine ring upon deprotonation. No correlation of these changes in coupling constant is observed with the pH dependence of chemical shift. Evidently s orbital changes are small compared to p orbital changes during deprotonation in the compounds.

As an aid to the assignment of the various resonances, two- and three-bond proton–carbon coupling constants were also determined. These are reported in Table V.

Acknowledgments. The authors gratefully acknowledge support of this work under Grant AM 15707 from the National Institutes of Health and GP-37025 from the National Science Foundation.

Registry No.—I, 17281-92-4; II, 54-47-7; III, 85-87-0, IV, 529-96-4; V, 65-23-6.

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The Chemistry of 2-Alkoxy-3,4-dihydro-2*H*-pyrans. III. Synthesis and Solvolysis of the Dichlorocarbene Adducts 3-Alkoxy-2-oxa-7,7-dichloronorcaranes

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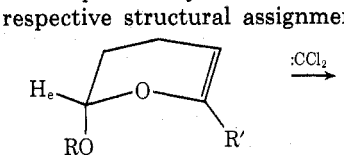
Received February 7, 1975

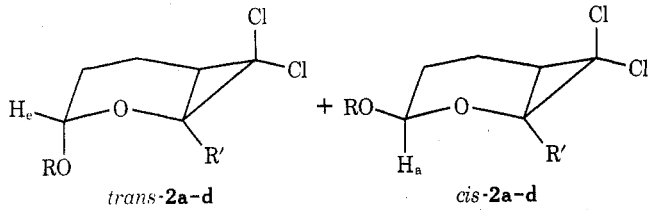
trans- and *cis*-3-alkoxy-2-oxa-7,7-dichloronorcaranes (**2a,b**) and *trans*- and *cis*-3-alkoxy-1-methyl-2-oxa-7,7-dichloronorcaranes (**2c,d**) were prepared by the addition of dichlorocarbene to 2-alkoxy-3,4-dihydro-2*H*-pyrans (**1a,b**) and 2-alkoxy-6-methyl-3,4-dihydro-2*H*-pyrans (**1c,d**), respectively. The addition, which is rather stereoselective owing primarily to the steric interactions of the axial 2-alkoxy group on the 3,4-dihydro-2*H*-pyran ring, yields predominantly the trans product. Subsequent solvolysis of the trans–*cis* mixtures **2a** and **2b** in alcoholic silver nitrate yielded 2-chloro-1,1,6,6-tetramethoxy-*cis*-2-hexene (**3a**) and 2-chloro-1,1,6,6-tetraethoxy-*cis*-2-hexene (**3b**), respectively. Similar treatment of **2c** and **2d** resulted in the formation of 3-chloro-7,7-dimethoxy-*cis*-3-hepten-2-one (**4a**) and 3-chloro-7,7-diethoxy-*cis*-3-hepten-2-one (**4b**), respectively. Evidence is presented that the electrocyclic ring opening requires the synchronous assistance of the equatorial 3-alkoxy substituent.

For some time we have been interested in the rather unusual effect of ring substituents on the chemistry of 3,4-dihydro-2*H*-pyrans.[2] In particular, an alkoxy group at the C-2 position seems to play a significant role in the outcome of electrophilic additions to the dihydropyran **1**.[2,3] We now describe the influence of the 2-alkoxy group on the addition of dichlorocarbene to the title compounds **1a–d**, and the subsequent solvolytic rearrangement studies of the dichlorocarbene adducts **2a–d** in alcoholic silver nitrate solutions.

Addition of dichlorocarbene, generated by the decomposition of ethyl trichloroacetate with sodium methoxide,[4] to 2-alkoxy-3,4-dihydro-2*H*-pyrans (**1a,b**)[5] and 2-alkoxy-6-methyl-3,4-dihydro-2*H*-pyrans (**1c,d**)[5] yielded a trans–cis mixture of the corresponding 3-alkoxy-2-oxa-7,7-dichloronorcaranes (**2a,b**) and 3-alkoxy-1-methyl-2-oxa-7,7-dichloronorcaranes (**2c,d**), respectively. The trans–cis mixtures were separated by careful column chromatography, and the respective structural assignments were based on spectral data and composition analyses. The stereochemical and conformational assignments of the adducts **2a–d** were made by analyzing the 100-MHz NMR spectra of the products (see Table I) and are consistent with the assigned conformation of the substituted dihydropyrans **1a–d**.

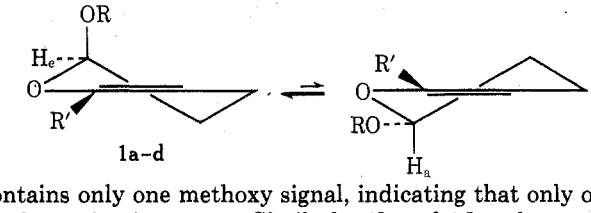


1a, R = CH_3; R′ = H
b, R = C_2H_5; R′ = H
c, R = CH_3; R′ = CH_3
d, R = C_2H_5; R′ = CH_3



trans-**2a–d** *cis*-**2a–d**

The conformations of the 2-alkoxy-3,4-dihydro-2*H*-pyrans (**1a–d**) were assigned by inspection of the 100-MHz NMR spectra. Two conformations for the 2-alkoxy-3,4-dihydro-2*H*-pyrans (**1a–d**) are possible, one with an equatorial anomeric proton (H_e) and another with an axial anomeric proton (H_a). The NMR spectrum of **1a** and **1c** each



1a–d

contains only one methoxy signal, indicating that only one conformation is present. Similarly, **1b** and **1d** each contain only one ethoxy triplet (see Table II). Since the anomeric proton of each 2-alkoxy-3,4-dihydro-2*H*-pyran (**1a–d**) is clearly a triplet, where $J_{ae} = J_{ee}$, the dihydropyrans **1a–d** exist predominantly (greater than 90%) in the conformation where the anomeric proton (H_e) is equatorial. Such a conformation is also predicted by the anomeric effect (Edward–Lemieux effect)[6] and makes the rather stereoselective addition of the dichlorocarbene to the olefins **1a–d** understandable.

The presence of a bulky axial group (the alkoxy substituent of the C-2 position) would result in a preferential trans addition to the dihydropyran **1**, yielding *trans*-**2** as the predominant product containing an anomeric equatorial proton (see Table I). Addition to the less favored sterically hindered side of the molecule would yield the minor product *cis*-**2**, which would assume a conformation containing