Unusual Coordination and Metal–Ligand Geometry of a Vanadyl Porphyrin in Aqueous Solution

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2 days needed to attain equilibrium. After P(3)P remained in the SDS and/or HTAC micellar solution for 2 days, all fluorescence behavior became the same as that for the corresponding frozen–thawed micelles.

In the present stage, the detailed mechanism for the freeze–thaw effect is not clear. A plausible mechanism is presented in the following. According to Menger's micelle model,[11] the micelle interiors of the ionic surfactants are considerably polar. P(3)P seems to be to hydrophobic to dissolve in micelles. At first, therefore, the surfactant micelles and the P(3)P aggregates coexist in the system. As an ice structure of the aqueous bulk phase grows, dehydration of the micelles may proceed leading to shrinkage of the micelles. The interior of the shrunken micelle should be so hydrophobic that P(3)P penetrates into the micelle core. P(3)P may dissolve in the micelle core by making spontaneously a hydrophobic environment in the vicinity of the P(3)P location site. Along with this dehydration mechanism, a concentration effect of freezing may account for the accelerated solubilization. Butler and Bruice have found fairly large acceleration of bimolecular reactions upon freezing.[12] They explained that reactants are concentrated in liquid regions between ice crystals to result in the rate enhancements. A similar effect may be considered for the solubilization of P(3)P in the micelles; i.e., both the micelles and P(3)P are concentrated in the liquid domains remaining between the ice crystals during freezing, which leads to the increase in the probability of the collision of P(3)P with the micelle. We believe that the concentration effect cooperates with the dehydration of micelles to accelerate the solubilization of P(3)P. It seems that the effect of the freezing and thawing on the solubilization rate can appear for the micelles whose interiors are considerably polar.

The present study not only revealed an interesting property of the surfactant micelles, which has not been known, but also suggested the possibility of the use of micelles as a simple model for investigating freezing injury of biological cells.

Registry No. P(3)P, 61549-24-4; SDS, 151-21-3; HTAC, 112-02-7; Triton X-100, 9002-93-1; Brij 58, 9004-95-9; Emulgen 911, 9016-45-9.

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Unusual Coordination and Metal–Ligand Geometry of a Vanadyl Porphyrin in Aqueous Solution

John A. Shelnutt* and Mary M. Dobry

Division of Chemical Physics of Organic Materials, Sandia National Laboratories, Albuquerque, New Mexico 87185
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Vanadyl uroporphyrin I in aqueous alkaline solution has considerably modified absorption and resonance Raman spectra when compared with vanadyl porphyrins in organic solvents or in the crystalline state. Under these latter conditions vanadyl porphyrins are known to contain 5-coordinate, out-of-plane vanadium(IV). The unusual spectra of vanadyl uroporphyrin in water can be converted to spectra typical of the 5-coordinate vanadyl porphyrins by formation of π–π complexes or by π–π dimerization. Because π–π complex formation and dimerization block addition of an axial sixth ligand a 6-coordinate, in-plane dihydroxide vanadium(IV) uroporphyrin complex is indicated. This hypothesis is consistent with an analysis of the frequencies of the core-size marker lines of metalloporphyrins indicating expansion of the metal core by 0.03–0.04 Å in $V(OH)_2$UroP.

Vanadyl porphyrins are widely distributed and naturally occurring components of sediments and petroleum deposits.[1] Such geoporphyrins are of biological origin, and their diagenesis provides a valuable molecular fossil record of past environmental conditions in these geological formations. Changes in the absorption spectra of metallopetroporphyrins resulting from formation of coordination complexes are useful for identification purposes.[2]

Two forms of vanadyl uroporphyrin I (VOUroP) are observed in aqueous alkaline solution as evidenced by the absorption spectra in Figure 1. In the presence of 1,10-phenanthroline (phen) the absorption spectrum is typical of pyrrole-substituted vanadyl porphyrins in organic solvent (CH_2Cl_2) which have an α-band near 572 nm, a β-band at 533 nm with about one-half the intensity of the α-band, and a Soret band near 407 nm.[3] Based on a comparison of their resonance Raman spectra, vanadyl porphyrins in organic solvents possess a metal–ligand geometry similar to the X-ray crystal structures.[4,5] The vanadium(IV) ion is 5-coordinate and out of the plane of the porphyrin ring by about 0.5 Å and the V=O bond length is 1.62 Å. The center-to-nitrogen distance (or core size) is 2.03 Å.[5] (1 in Figure 2.)

In the absence of added phen, however, an aqueous alkaline solution of VOUroP has an unusual visible absorption spectrum with inverted α/β band intensity ratio. Also as can be seen in Figure 1 all bands are strongly red shifted with the α-band at 584 nm and the Soret at 423 nm. The band at 336 nm also gains considerable intensity

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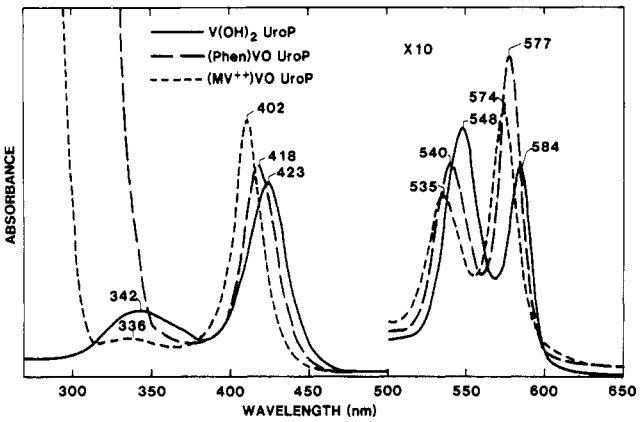

Figure 1. Absorption spectra of vanadyl uroporphyrin I in 0.1 M NaOH (—) and the $\pi-\pi$ complexes with 1,10-phenanthroline (phen) (– – –) and methyl viologen (MV^{2+}) (----). The porphyrin concentration is 1.7×10^{-5} M for all three spectra.

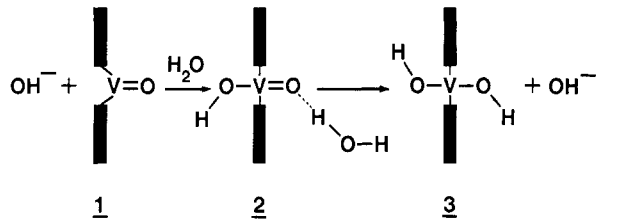

Figure 2. Formation of vanadium(IV) uroporphyrin dihydroxide from vanadyl uroporphyrin.

and shifts to 342 nm. Formation of the 6-coordinate dihydroxide species ($V(OH)_2$UroP) **3** in Figure 2 via intermediate **2** is suggested and is a consequence of the water solubility of vanadyl uroporphyrin.

Support for the formation of the dihydroxide is given by a comparison of the absorption and resonance Raman data under various conditions in which the sixth axial ligand site is blocked. For example, upon binding phenanthroline, a $\pi-\pi$ charge-transfer complexing agent,[6-9] or upon dimerization by addition of salt (5.5 M),[10-13] a near normal visible spectrum is observed (Figure 1) with the α-band at 577 nm only slightly red shifted with respect to the spectrum of vanadyl porphyrins in organic solvents. The Soret band of the phen–VOUroP complex at 418 nm is closer to the usual vanadyl porphyrin absorption. The dimer species also shows the normal α/β band ratio, but the Soret band exhibits a large blue shift to 397 nm and reduced absorbance typical of aggregated metallouroporphyrins.[10-13]

Similarly, all resonance Raman spectra of vanadium uroporphyrins are typical except for the monomeric V-$(OH)_2$UroP species. The resonance Raman spectrum in 0.1 M NaOH is shown in Figure 3 along with that of the phen–VOUroP complex. The spectra were taken simultaneously with the RDS apparatus described previously.[7] With 514.5-nm excitation several marker lines of porphyrin structure are strongly enhanced. The Raman line of VOUroP at 1380 cm^{-1} (ν_4) is the oxidation-state marker line of iron porphyrins and heme proteins.[14-15] Also the

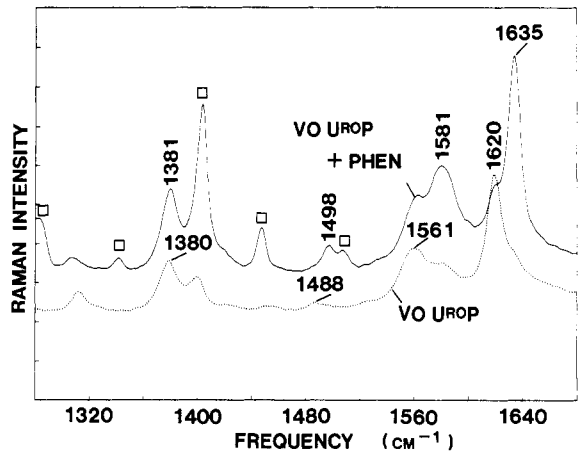

Figure 3. Resonance Raman spectra of vanadyl uroporphyrin I in 0.1 M NaOH (····) and the $\pi-\pi$ complex with 1,10-phenanthroline (—) showing the high-frequency marker line region. [VOUroP] = 1×10^{-4} M. Laser excitation is at 514.5 nm (600 mW). Spectral resolution is 3 cm^{-1}. The squares indicate unbound phen lines. Phen is at saturation.

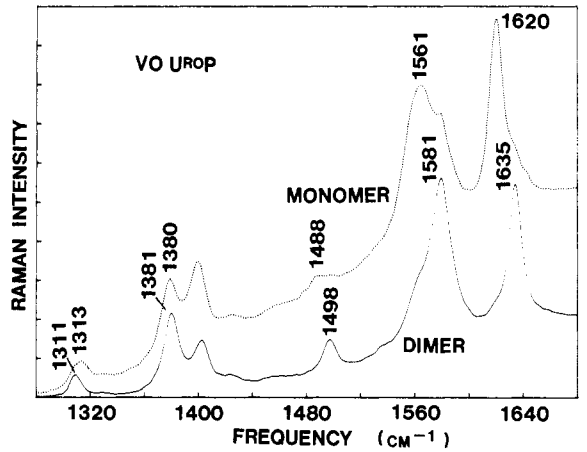

Figure 4. Resonance Raman spectra of vanadyl uroporphyrin I in 0.1 M NaOH (····) and the salt-induced dimer (—). Laser excitation is at 528.7 nm (200 mW). Spectral resolution is 3 cm^{-1}. NaCl is near saturation.

vibrations at 1488 (ν_3), 1561 (ν_{19}), and 1620 cm^{-1} (ν_{10}) are well-known markers of the porphyrin core size.[16] For comparison, VO etioporphyrin (VOEtioP) in a KBr matrix, for which a 5-coordinate crystal structure is known,[5] exhibits lines at 1380 (ν_4), 1500 (ν_3), 1574 (ν_{19}), and 1635 cm^{-1} (ν_{10}).[16,17] The core-size marker lines of crystalline VOEtioP are much higher (about 13 cm^{-1}) than those of V-$(OH)_2$UroP.

On the other hand, the phen–VOUroP complex gives lines at 1381, 1498, 1581, and 1635 cm^{-1} that are closer to VOEtioP frequencies. Because phenanthroline-complex formation is not complete, lines from the uncomplexed aqueous form are also evident in the spectrum. In addition a small amount of a form similar to the phen complex is evident in the solution of VOUroP without added phenanthroline.

Aggregation of uroporphyrins, which results from neutralization of the eight negatively charged acid groups at the periphery by salt cations, also blocks addition of a sixth ligand. In Figure 4 the resonance Raman spectra of VOUroP monomer and salt-induced dimer are compared.

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The monomer spectrum, taken with 528.7-nm excitation, is generally as described above. For the dimer the lines are close to frequencies of the phen–VOUroP complex and crystalline VOEtioP. Dimerization is almost complete at 5.5 M NaCl as lines from the uncomplexed monomeric form are almost absent.

Dimerization of VOUroP also occurs at acid pH and gives absorption and resonance Raman spectra similar to the salt-induced dimer, i.e., normal α/β ratio and frequencies of core-size and oxidation-state marker lines near those of the salt-induced dimer. The pK of 6.7 for acid dimer formation is the same as for copper uroporphyrin. However, the in-plane copper uroporphyrin forms polymers and precipitates, whereas VOUroP, which cannot form units larger than dimers because of steric hindrance of the vanadyl oxygens, does not.

These Raman results further support the formation of a 6-coordinate dihydroxide vanadium porphyrin complex, because an anomalous Raman spectrum is observed only when the sixth ligand site is unblocked. The shifts in the core-size marker lines further suggest that the dihydroxide monomeric form is an in-plane species (**3** in Figure 2).

First, from the correlation between ν_{10} and core size that is given in Spaulding et al.[16] a 1-cm^{-1} decrease in ν_{10} represents about 0.0024-Å increase in the center-to-nitrogen (pyrrole) distance. Therefore the porphyrin core is expanded in the dihydroxy species by 0.035 Å relative to the dimer and phen complex. In the 5-coordinate vanadyl octaethylporphyrin crystal structure,[5] the center-to-nitrogen distance is 2.031 Å. The frequency of ν_{10} for the crystal (1635 cm^{-1}) is identical with that of the phen complex and dimer; therefore, the core expands to 2.066 Å in the dihydroxy complex. An increase in core size to 2.066 Å would allow the vanadium ion to move from 0.54 Å out of the plane of the pyrrole nitrogens to only 0.38 Å out-of-plane, assuming the V–N distance is held constant at 2.102 Å. Slightly different numbers are obtained by using ν_3 and ν_{19}, but it is clear that the core expands by about 0.03 Å.

In addition, a decrease in the V–N distance of only 0.03 Å to 2.07 Å would allow the vanadium to be in-plane in an expanded porphyrin ring. This V–N distance is intermediate between the unequal V–N bond lengths found in the asymmetric deoxophylloerythroetioporphyrin.[4] In this structure the V–N distances range from 2.13 to 1.96 Å for the pyrrole with the fused ring. The V–N distance 2.053 Å is found in *N,N'*-ethylenebis(acetylacetoneiminato)oxovanadium(IV).[18]

Nonbonding repulsive interaction between the oxygen and the pyrrole nitrogens determine the out-of-plane position of the vanadium ion because the V=O bond is strong and the bond length relatively constant.[4,20] Vanadium is probably not out-of-plane because of its size, since its ionic radius (0.59 Å for V(IV))[19] is smaller than for some in-plane metal–porphyrin complexes such as Pt and Cu.

Formation of the dihydroxide species **3** as shown in Figure 2 explains the structural changes indicated by the Raman results. Binding OH$^-$ to the positively charged vanadium[20] and hydrogen bonding to the vanadyl oxygen allow formation of the V(IV) 6-coordinate species as shown. The longer V–OH bonds allow the metal to move in-plane at the expense of a small expansion in the porphyrinato core. Increased nonbonding repulsion between hydroxyl oxygens and pyrrole nitrogens and movement of the metal into the porphyrin plane accounts for the expanded core. Also, hydrogen bonding of the hydroxides to water molecules may confer some double bond character to the V–O bonds.

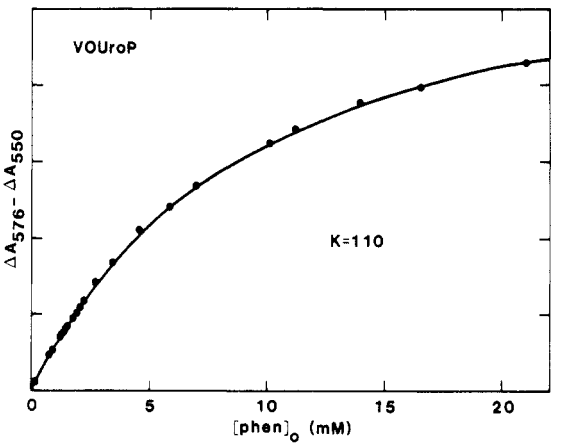

Figure 5. Equilibrium binding data for the phen–vanadyl uroporphyrin complex. Percent binding is determined from the difference in minimum and maximum absorbance in the difference spectrum. The peak and valley in the difference spectrum result from the complex-induced blue shift in the α-band. The theoretical curve assumes a 1:1 binding equilibrium, phen + porphyrin $\rightleftarrows$ phen–porphyrin; K = 110 from the least-squares fit to the data points.

Hydrogen bonding to the vanadyl oxygen without OH$^-$ binding to vanadium might explain the structural change in the porphyrin core, but cannot account for the effects of π–π complexation and dimerization. Figure 5 shows the equilibrium binding data for the phenanthroline complex with VOUroP. The theoretical curve assumes a 1:1 complex, and is seen to give an adequate least-squares fit to the data (K = 110, 25 °C). For comparison analysis of binding data for the phen–CuUroP complex requires formation of a 2:1 complex (K_1 = 7100, K_2 = 320; 27°C).[7,8] For VOUroP, addition of the second phen is blocked by strong axial ligation of the vandyl oxygen. It is also apparent that the presence of bound OH$^-$ greatly lowers the affinity of VOUroP for even one phen in relation to the CuUroP complex. (Compare K's given above.) Therefore, phen binding blocks the back-side vanadium binding site, displacing OH$^-$, but probably does not affect the vanadyl oxygen site.

An aromatic cation such as methyl viologen (MV^{2+}) is "clamped" across the porphyrin ring by electrostatic attraction between the positive charges at each end of viologen and ionized carboxylate groups on opposite edges of the porphyrin ring.[7-9] Therefore, the sixth ligand site is covered by the aromatic rings of the viologen molecule and OH$^-$ binding is again blocked. Consequently, we again expect formation of a 5-coordinate species. The core-size marker lines at 1496 (ν_3), 1576 (ν_{19}), and 1632 cm^{-1} (ν_{10}) are consistent with a 5-coordinate form, but are 2–5 cm^{-1} lower than for the phen complex or dimer as was also observed for viologen complexes with other metalloporphyrins.[6-9] The Raman lines are low for the viologen complex as a result of charge acceptance by the porphyrin. In contrast the porphyrin is a donor in phen complexes. The shift in the oxidation-state marker line of the viologen complex by −3 cm^{-1} relative to the phen–VOUroP complex and the dimer is also consistent with other metallouroporphyrin–methyl viologen complexes.[6-9] Because viologen complexation usually has less of an effect on the visible absorption spectrum of metalloporphyrins than dimerization or phen binding it should give an α-band maximum closest to that observed in organic solvents[3] and this is the case as can be seen in Figure 1.

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Gouterman has predicted vanadyl porphyrins would have the vanadium atom out of plane because molecular orbital calculations for an in-plane vanadium gave strong ultraviolet absorption bands characteristic of hyperporphyrins, and no such bands had been observed. The strong UV band near 342 nm and the red shift of the $\pi \rightarrow \pi^*$ bands of $V(OH)_2UroP$ are consistent with the prediction of the MO calculations for an in-plane vanadium(IV) porphyrin (M. Gouterman, personal communication).

In previous work on VO-porphyrin complexes with possible ligands,[21] β-carbon substituted vanadyl porphyrins have been shown to form weak (K = 0.02, 25 °C) complexes with nitrogen bases such as pyridine in organic solvents. However, no inversion of α/β band intensity ratio is observed and shifts in the absorption maxima are small. Pyridine may complex with the ring in these complexes as has been observed for other porphyrins.[8,12,22]

In contrast, in earlier work on meso-substituted vanadyl porphyrins large red shifts on binding piperidine were observed.[23] Binding is again weak (K = 0.4). The spectral shifts were thought to indicate formation of a 6-coordinate species, and this may be the case, since the α/β band ratio is strongly affected. Thus, the piperidine complex may be similar to the vanadium porphyrin dihydroxide complex reported here.

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Structure and Dynamics of Molecular Rotation of 2-Pyridone in Toluene and Methanol by Carbon-13 Nuclear Magnetic Resonance

J. Guilleréz, B. Tiffon, B. Ancian, J. Aubard, and J. E. Dubois*

Institut de Topologie et de Dynamique des Systèmes de l'Université Paris VII, associé au C.N.R.S., 75005 Paris, France
(Received: April 27, 1983)

The structure and dynamics of molecular rotation of 2(1*H*)-pyridone and 1-methyl-2(1*H*)-pyridone in toluene and methanol have been investigated at 305 K by ^{13}C NMR. Both chemical shifts and relaxation times show that 2-pyridone forms stable hydrogen-bonded complexes in methanol, reorienting as a proper unit taking with it two solvent molecules. It follows that the lifetimes of these complexes are greater than the rotational correlation time scale—10^{-11}–10^{-12} s.

Due to the role of hydrogen bonding in biochemical processes involving nucleic bases and proteins, there is a continuing interest in understanding the solvation of the heterocyclic amide group. Recent theoretical[1] and experimental studies by ^{17}O NMR[2] and calorimetry[3] have confirmed that the carbonyl oxygens of oxopyrimidines are proton acceptors in hydroxylic solvents. Estimation of the lifetimes of the complexed species is then of prime importance since specific recognition of nucleic bases through hydrogen bonding by proteins would implicate desolvation as a preliminary step. In this context, nuclear magnetic relaxation measurements have shown that the pyridine–water and pyridine–methanol complexes are stable within the range of the rotational correlation times[4] and reorient as proper units at ambient temperature. However, it was very recently shown that saturated cyclic lactams do not exhibit significant solvation effects in water: the monomer seems to be the only species which reorients in this medium while dynamics in inert solvent is consistent with dimerization.[5]

In this communication, we report some preliminary carbon-13 NMR results on the structural and dynamical properties of 2-pyridone in toluene and methanol. This substrate, which is commonly used as a model for pyrimidinic nucleic bases, is known to be strongly solvated in hydroxylic solvents.[6-8] Experiments were performed at 303 K in the Fourier transfer mode on either a Bruker WP 200 or WH 400 spectrometer under proton broad band decoupling conditions. Degassed 0.1 M solutions of purified commercial 2(1*H*)-pyridone (sublimed) and 1-methyl-2(1*H*)-pyridone (vacuum distilled) were contained in 10 mm o.d. tubes. Field-frequency stabilization was achieved by using the CD_3 signal of small added quantities of perdeuterated solvent. Carbon resonances were assigned by using the gated-coupling mode and agree with previ-

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