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On the Reactivity of Phosphate Monoester Dianions in Aqueous Solution: Brønsted Linear Free-Energy Relationships Do Not Have an Unique Mechanistic Interpretation

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Phosphoryl transfer plays a fundamental role in many important functions of the cell. For this reaction to occur, the P−O bond of the leaving group has to be broken, and a new P−O bond must be formed between the phosphorus and the attacking group

$$R'OH + R''OPO_3^{2-} \rightleftharpoons R'OPO_3^{2-} + R''OH \qquad (1)$$

Depending on the order of the bond-breaking and bond-forming events, actual reaction mechanisms should, in principle, fall between two limiting cases of the associative and dissociative mechanisms.[1−5]

Several experimental techniques are available to probe the character of the transition state involved.[6] Among them, the observed linear free-energy relationships (LFER) between the activation and equilibrium free energies have been frequently used. In particular, the rate constant (k) for the uncatalyzed reaction 1 in aqueous solution has been found by Kirby, Varvoglis, and Jencks to be highly sensitive to the dissociation constant of the leaving group (R″OH),[7]

$$\log(k) = 0.86 - 1.23\mathrm{p}K_a(R''OH) \qquad (2)$$

but practically independent of the pK_a of the nucleophile (R′OH)[8]

$$\Delta\log(k) = 0.13\Delta\mathrm{p}K_a(R'OH) \qquad (3)$$

The measured Brønsted coefficients $\beta_{lg} = -1.23$ and $\beta_{nuc} = 0.13$ were interpreted as the evidence that reaction 1 proceeds via a dissociative mechanism, in which the anion of monomeric metaphosphate (PO_3^-) is formed.[7,9] However, this interpretation was based on an assumption that the associative mechanism, characterized by the formation of the 5-coordinated intermediate or transition state, is inconsistent with the experimental LFER.[10,11] In this paper, we address the validity of this assumption by ab initio calculations of the kinetic substituent effects for the phosphoryl transfer reaction involving the 5-coordinated transition state. This study is a continuation of our effort to compare the energetics of the associative and dissociative pathways of phosphate monoester hydrolysis and to scrutinize the interpretation of experimental data on this important reaction.[5,12]

In the first part of our study we evaluated a section of the free energy surface for the nucleophilic attack of methanol on phenyl phosphate dianion in aqueous solution. These calculations involved the HF/6-31G* partial geometry optimizations, in which the P−OMe and P−OPh bond lengths were varied by 0.1 Å increments in the 2.3−1.7 Å and 2.5−1.7 Å range, respectively. In this free energy region, the proton was found to be fully transferred to the equatorial phosphate oxygen and oriented toward the methoxide group. For each of the resulting 63 structures, we evaluated the MP2/6-31+G** gas-phase energy (ΔE_{gas}) and solvation free energy ($\Delta\Delta G_{solv}$) The solvation calculations were carried out using polarized continuum (PCM/HF/6-31G*)[13] and iterative Langevin dipoles (LD)[14,15] models (for details of the computational protocol see refs 5 and 14). The two different computational methods were applied to reveal possible errors related to the approximative nature of these solvation models (for example, neither of these models treats rigorously the first solvation-shell effects). The free energy profile (potential of mean force) was calculated as

$$\Delta g = \Delta E_{gas} + \alpha\Delta S_{gas} + \Delta\Delta G_{solv} \qquad (4)$$

where the H-bonded complexes, in which the P−O(nuc) and P−O(lg) distances were fixed at 3.0 and 1.75 Å, were used as the reference states. Note that the distance of 1.75 Å was found by us to correspond to the equilibrium P−O(lg) bond length for phosphate monoester dianions in aqueous solution (Figures 1S and 2S). Consistent with our previous studies,[5,12] the coefficient α in eq 4 was chosen as zero.[16]

The calculated relative free energies Δg (Figure 1) indicate that there is a minimum on the free energy surface, which corresponds to the dianionic pentacovalent intermediate (phosphorane). Previously, the existence of the dianionic phosphorane intermediate has been the subject of several theoretical studies.[17−19] The first of these studies established the nonexistence of this intermediate in the gas phase.[17] Later, it was pointed out that the dianionic phosphorane becomes marginally stable upon inclusion of one or more explicit water molecules in the calculation.[18,19] Clearly, the stability of the phosphorane minimum is the consequence of the fact that the hydration energy favors smaller ions. Here, the size-dependence of the solvation free energy was found to be more pronounced for the PCM than for the LD solvation model.

The phosphorane intermediate is separated from the reactants and products by the transition states TS1 (MeO−P = 2.15−2.20 Å, P−OPh = 1.8−1.85 Å), and TS2 (MeO−P = 1.8 Å, P−OPh = 2.20 Å). In both of the TS1 and TS2 structures, the hydrogen

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(10) The nature of the observed LFER depends strongly on the shape of the relevant potential energy surface and its decomposition into intersecting parabolas (see, e.g., ref 11). If this surface can be represented by only two intersecting parabolas, the resulting LFER are expected to provide conclusive mechanistic information. Unfortunately, this may not be the case if the potential surface involves several transition states.)
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(16) To support the neglect of the entropic term, we evaluated this term for TS2 structure using ideal-gas, rigid rotor, and harmonic (HF/6-31G* frequencies) approximations. This calculation yielded $\Delta S_{gas}^{\ddagger}$ in the 4.0 ± 0.2 eu range for all leaving groups. Thus, the entropy-related substituent effects fall below 0.1 kcal/mol. Also note that the overall entropy contribution is quite small and originates entirely from the vibrational degrees of freedom.
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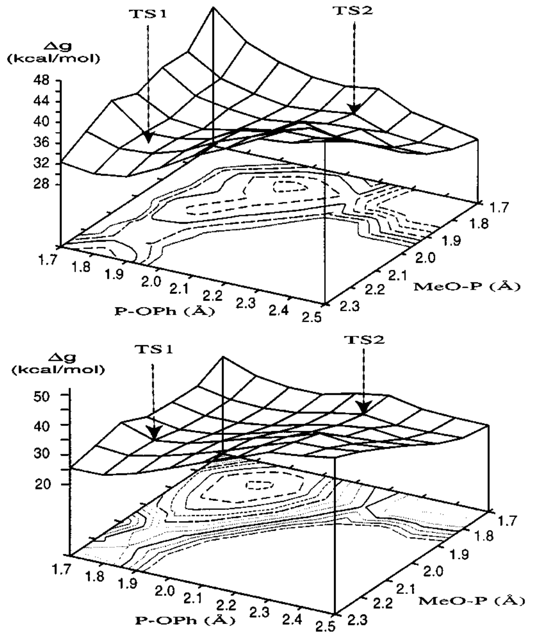

Figure 1. Section of the free-energy surface for the methanolysis of phenyl phosphate dianion calculated using approximation 4. The Δg values for the phosphorane intermediate obtained at the MP2+LD and MP2+PCM levels amount to 31.3 and 22.9 kcal/mol, respectively. The isoenergetic levels are drawn with the resolution of 0.7 kcal/mol. Top: MP2/6-31+G** + LD results. Bottom: MP2/6-31+G** + PCM results.

atom bonded to the equatorial oxygen was found to be oriented toward the nucleophilic oxygen. Because the stabilizing electrostatic effect of this hydrogen on the TS1 structure is balanced by the larger stability of the leaving PhO^- group in the TS2 structure, the energies of TS1 and TS2 are nearly the same.

To examine the LFER that could be expected if reaching TS2 was the rate-limiting step in the associative reaction mechanism, we examined the effects of the chemical modification of the attacking and leaving group on the relative energy of the TS2 structure in aqueous solution. Here, the P−O(nuc) and P−O(lg) distances were kept at 1.8 and 2.2 Å,[20] respectively, and the remaining degrees of freedom were relaxed at the HF/6-31G* level. However, this relaxation did not change the position of the equatorial H, which remained oriented toward the nucleophile.

We found that the gas-phase activation energies decrease for larger and more polarizable leaving groups (Table 1). This trend occurs at both HF and MP2 levels of theory. Also, it seems to be somewhat sensitive to the basis set extension. Although the overall solute size becomes smaller upon going from the hydrogen bonded complex to the TS2 structure, solvation contributions were found to increase the activation barrier. This is because the charge of the solute becomes more delocalized in the transition state. The substituent effects on the gas phase and solvation energies oppose each other. However, this compensation is not complete so that there is a significant substituent effect on the total activation free energy $\Delta g^{\ddagger}$. The plot of the calculated first-order rate constants [21] versus pK_a of the leaving group (Figure 2) demonstrates that the LFER predicted for the associative mechanism are consistent with the experimental LFER (eq 2). In addition, our calculations for the attack of trifluoro ethanol (pK_a = 12.4) and methanol (pK_a = 15.5) on phenyl phosphate dianion (Table 1) show that the basicity of the nucleophile has little effect on the activation barrier corresponding to the TS2 structure. This result is in agreement with the experimental findings.[7,22]

Table 1. Substituent Effects on the Relative Energy and Relative Solvation Free Energy of the TS2 Structure

		$\Delta E_{gas}^{\ddagger b}$		$\Delta\Delta G_{solv}^{\ddagger c}$	
nucleophile	leaving group[a]	HF	MP2	LD	PCM
CH_3OH	CH_3OH	45.6	39.9	3.0	−3.8
CH_3OH	Ph	25.5	23.3	8.8	4.6
CH_3OH	*o*-ClPh	20.1	18.6	11.7	7.4
CH_3OH	*o*,*p*-ClPh	17.4	17.1	11.2	9.4
CH_3OH	*o*-Cl, *p*-NO_2Ph	8.6	8.7	13.5	12.1
CF_3CH_2OH	Ph	25.1	23.1	9.2	7.3

[a] Phenol is abbreviated as Ph. Prefixes *o* and *p* denote substitions in the positions ortho and para with respect to the bridging oxygen. [b] Activation energy (kcal/mol) calculated as the difference in the total energies of TS2 and the corresponding reference state (see the text). HF denotes the HF/6-31G* method. MP2 denotes the MP2/6-31+G**//HF/6-31G* method. [c] The difference in the solvation free energies (kcal/mol) of TS2 and the corresponding reference state. LD denotes the iterative Langevin dipoles calculation.[14] [e] PCM denotes PCM/HF/6-31G* calculation using Pauling's atomic radii scaled by the factor 1.2.

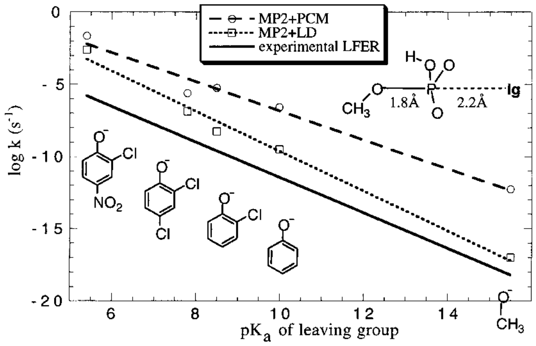

Figure 2. Comparison of the experimental (eq 2) and calculated LFER. The experimental LFER was observed in the pH-independent region above pH 7.[7] The Brønsted slopes (β_{lg}) fitted to the calculated rate constants amount to −1.38 and −1.01 for MP2+LD and MP2+PCM data, respectively. Values of pK_a constants were taken from ref 24.

In conclusion, the results presented in this paper do not support the generally accepted view that the LFERs (eqs 2, 3) observed by Kirby and Varvoglis[7] represent conclusive evidence for the dissociative mechanism of the hydrolysis of phosphate ester dianions.[2,4,7] This does not mean that the observed LFER should be interpreted as evidence for the associative mechanism but rather that the interpretation of this relationship is not unique. In fact, the LFER appear to correspond to a late TS in the associative mechanism[23] as well as to an early transition state in the dissociative mechanism, since both mechanisms involve a largely broken bond between the phosphorus and the leaving group oxygen.

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Supporting Information Available: Calculated substituent effects on the equilibrium P−O(aryl) bond lengths in aryl phosphate dianions in aqueous solution (3 pages, print/PDF). See any current masthead page for ordering information and Web access instructions.

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(20) The use of the fixed bond length to the nucleophile and to the leaving group is justified by the flat free energy surface in the vicinity of TS2 and by the fact that the substituent effects on the P−OX (X = aryl) bond length of dianions of phosphate monoesters were found to be very small (Figures 1S and 2S).

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(23) It should be noted that the experimental LFER can be reproduced for the associative mechanism only if TS2 has higher energy than TS1. On the basis of the present results, this condition is satisfied if the difference in the pK_a constants of the attacking and leaving groups is smaller than 6. However, one should realize that the error of the calculated relative free energies of TS1 and TS2 is likely to be larger than the error of the predicted substituent effects. This is because the orientation of the phosphate hydroxyl group induces a significant structural difference between TS1 and TS2. Consequently, it is possible that the TS2 structure could correspond to the rate-limiting step of associative hydrolysis even for good leaving groups.

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