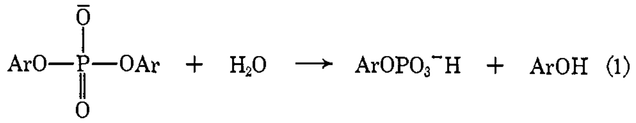
Micellar Effects upon the Hydrolysis of Bis-2,4-dinitrophenyl Phosphate[1]

G. J. Buist, C. A. Bunton,[2] L. Robinson, L. Sepulveda,[3] and M. Stam

Contribution from the Department of Chemistry, University of California, Santa Barbara, California 93106, and the Faculty of Chemistry and Pharmacy, University of Chile, Santiago, Chile. *Received December 10, 1969*

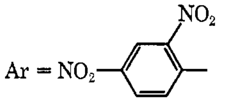
Abstract: Micellar effects upon the reaction between hydroxide ion and bis-2,4-dinitrophenyl phosphate to give 2,4-dinitrophenyl phosphate have been examined. The reaction is catalyzed up to 30-fold by cationic micelles of cetyltrimethylammonium bromide, CTABr, unaffected by anionic micelles of sodium lauryl sulfate, NaLS, and inhibited by uncharged micelles of a polyether. Added salts, particularly those with large, low charge density anions effectively inhibit catalysis by CTABr. Turbidity measurements show that the substrate and CTABr form submicellar aggregates, but that added salts reduce their formation. The ability of salts to inhibit formation of these submicellar aggregates increases with decreasing charge density of the anion, *e.g.*, tosylates are more effective than chlorides. The micellar effects upon the second stage of the reaction, hydrolysis of 2,4-dinitrophenyl phosphate, follow the usual pattern of catalysis by CTABr and no effect of an anionic or uncharged detergent and only a slight rate enhancement by hydroxide ion. The catalysis of the hydrolysis of 2,4-dinitrophenyl phosphate by CTABr and CTACl is not sensitive to large changes in detergent concentration which lead to a change in micellar shapes.

The spontaneous hydrolysis of bis-2,4-dinitrophenyl phosphate monoanion is a relatively slow reaction which involves rate-limiting attack of water upon the monoanion[4] (eq 1), whereas the faster hydrolysis of 2,4-dinitrophenyl phosphate dianion involves a rate-limiting heterolysis (eq 2).[5,6]



$$\mathrm{ArO{-}\overset{\bar{O}}{\underset{O}{P}}{-}OAr + H_2O \longrightarrow ArOPO_3^{-}H + ArOH} \qquad (1)$$

$$\mathrm{ArO{-}PO_3^{2-} \longrightarrow Ar\bar{O} + PO_3^{-}} \qquad (2)$$



At higher pH hydroxide ion attacks the bis-2,4-dinitrophenyl phosphate monoanion,[4] but the corresponding reaction of 2,4-dinitrophenyl phosphate dianion is slower and makes less contribution to the overall reaction.[6] The spontaneous hydrolyses of 2,4- and 2,6-dinitrophenyl phosphate dianions are catalyzed by cationic micelles of cetyltrimethylammonium bromide, CTABr, but there is almost no catalysis of the reaction with hydroxide ion.[7]

The spontaneous hydrolyses of 2,4-dinitro- and bis-2,4-dinitrophenyl phosphates occur with complete P–O fission, and predominant P–O fission is observed in the reaction of hydroxide ion with bis-2,4-dinitrophenyl phosphate.[4,8]

Because of the observation that biologically important phosphorylations can involve formally negatively charged species as the phosphorylating agents (for discussion of chemical models see ref 9 and 10), we were interested in studying micellar effects involving nucleophilic attack upon the anion of a phosphate ester, in order to extend existing observations of micellar effects upon nucleophilic attack on phosphoryl phosphorus.[11] We were also interested in finding whether anionic or uncharged micelles had any kinetic effect.

Experimental Section

Materials. The preparation and purification of the reagents have been described.[4,7] The cationic detergents were cetyltrimethylammonium bromide and chloride (CTABr and CTACl), the anionic detergent was sodium lauryl sulfate (NaLS), and the uncharged one was Igepal 24, which is a dinonyl phenol condensed with 24 ethylene oxide units, and which we designate DNPE. The dioxane was purified by the standard method of treating it with HCl, then with water and NaOH, and it was finally refluxed and distilled over molten sodium.

Kinetics. The hydrolysis was followed spectrophotometrically using a Gilford spectrophotometer with a water-jacketed cell compartment. In the first experiments we dissolved the substrate in a small amount of dioxane and added it as a dioxane solution. Unfortunately, even a small amount of dioxane affects the micellar catalysis, and therefore for most of the reactions we dissolved the substrate in water. Cordes and his coworkers have noted that micellar effects are very sensitive to adventitious impurities.[12] The absorbance of the 2,4-dinitrophenoxide ion was measured at 3580 Å.

The overall reaction is a two-step process where k_1 and k_2 are

$$\mathrm{(ArO)_2PO_2^-} \xrightarrow[k_1]{\mathrm{H_2O}} \mathrm{ArO^- + ArOPO_3^{2-}} \xrightarrow[k_2]{} \mathrm{ArO^- + H_2PO_4^-}$$

the first-order rate constants with respect to the phosphate esters. At pH < 10, $k_2 \gg k_1$ and the formation of 2,4-dinitrophenol has a

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[2] To whom inquiries should be addressed.

[3] University of Chile–University of California Cooperative Program Fellow on leave from the Faculty of Chemistry and Pharmacy, University of Chile, Santiago.

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[7] C. A. Bunton, E. J. Fendler, L. Sepulveda, and K.-U. Yang *ibid.*, **90**, 5512 (1968).

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[11] C. A. Bunton and L. Robinson, *J. Org. Chem.*, **34**, 773 (1969); C. A. Bunton, L. Robinson, and L. Sepulveda, *ibid.*, **35**, 108 (1970); *J. Amer. Chem. Soc.*, **91**, 4813 (1969).

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first-order kinetic form,[4] but at higher pH the system has to be treated as two consecutive first-order reactions, and the integrated rate equation can be solved using a simple program and the Culler-Fried on-line system connected to an IBM 360-75 computer.[13] In this system the information is displayed on the screen of a cathode-ray tube.

The integrated rate equation for consecutive first-order reactions can be written as

$$D_t = D_\infty \left\{ \frac{1 + (2k_2 - k_1)e^{-k_1t} - k_1e^{-k_2t}}{2(k_2 - k_1)} \right\} \qquad \text{(I)}$$

where D_t and D_∞ are the absorbances at times t and ∞. The computer was programmed to calculate D_t from the known value of D_∞ and assumed values of k_1 and k_2. The values of D_t(calcd) − D_t(obsd) were then displayed graphically, and the values of k_1 and k_2 were adjusted so that there was no systematic deviation of D_t(calcd) − D_t(obsd) in the course of the run. In practice k_2 was known approximately from earlier work[6,7] and this trial and error procedure took little time. Equation I assumes that the reaction starts at $t = 0$, and to allow for errors in the starting time, or the presence of products in the starting material, we introduced a correcting term δt to allow for these uncertainties and replaced t by $t + \delta t$, with δt being adjusted to give the best fit. The values of δt were always very small compared with the half-life of the overall reaction. Because of initial hydrolysis δt was larger when the substrate was dissolved initially in water rather than dioxane. As an objective test of the method the maximum, δ, and the standard deviation, σ, of D_t(calcd) from D_t(obsd) were calculated for each run, and were always in the range 0.003-0.008. This procedure would be unsatisfactory when $k_1 \sim k_2$, but this condition was not met in our reactions. In most experiments k_1 was considerably larger than k_2 and the best values of k_1 were obtained by concentrating the readings in the first 60% of reaction; this procedure meant that the values of k_2 were correspondingly less accurate, although this problem was of no great concern to us because they can be obtained directly.[7] Duplicate values of k_1 were within 5% of each other, but the corresponding figure was *ca.* 10% for k_2.

Hydrolyses of 2,4-dinitrophenyl phosphate at high detergent concentrations were followed directly.[7]

For reactions at low pH the values of k_1 were calculated graphically using the integrated first-order rate equation, because $k_2 \gg k_1$. All the rate constants are at 25.0°, and are first order with respect to phosphate ester, with units of sec^{-1}.

Most of the attack of hydroxide is upon phosphoryl phosphorus in the absence of detergents,[4] but because of the very low substrate concentrations used in the detergent-catalyzed reactions we could not determine the position of bond fission in this system. Evidence for P–O fission in the micellar-catalyzed reaction is that the maximum first-order rate constant for attack of OH$^-$ in the micellar phase upon the 2,4-dinitrophenyl group of 2,4-dinitrochlorobenzene in CTABr is *ca.* 10^{-4} sec^{-1} in 0.01 *M* NaOH,[14] and slower than the reaction in the micellar phase of bis-2,4-dinitrophenyl phosphate 0.01 *M* OH$^-$ in CTABr.

Results

Kinetics. The values of k_1 and k_2 were determined using hydroxide ion concentrations 0.01–0.1 *M* in the presence of CTABr (Tables I and II, Figure 1). Addition of 1.5 vol % dioxane approximately doubles both k_1 and k_2 in the presence of CTABr. Part of the effect could be caused by the ability of nonpolar solvents such as dioxane to reduce the critical micelle concentration (cmc) and so allow formation of micelles at low concentrations of detergent,[15] but this effect should not affect the maximum values of k_1 and k_2 in the "plateau" region where all the substrate is incorporated into micelles. Therefore we must assume that dioxane enhances the catalytic properties of CTABr micelles, possibly by decreasing their polarity. Spontaneous hydrolyses of dinitrophenyl phosphate dianions are assisted strongly by the addition of organic solvents to water,[5,6] but the effects are less marked for the reaction of the bis-2,4-dinitrophenyl phosphate monoanion.[4] However, the effects which we observe must be related to stabilization of the transition states relative to the initial states in the micellar phase, as well as to a change in the number of micelles present at a given detergent concentration.

Table I. Micellar Catalysis of the Reaction of Bis-2,4-dinitrophenyl Phosphate with Hydroxide Ion[a]

	10^3C_{CTABr}, *M*				
C_{OH}, *M*	0.50	1.00	2.00	3.00	4.00
0.005			0.46		0.46
0.010	0.23	0.59	0.87	0.85	0.94
0.010[b]	0.47	1.18	1.95	2.00	2.10
0.020	0.33	0.83	1.75	2.00	2.10
0.100		2.50	3.33	3.35	3.00

[a] Values of 10^3k_1, sec^{-1} in aqueous solution at 25.0°; in the absence of detergent, $k_1 = 0.03 \times 10^{-3}$ sec^{-1}, with 0.01 *M* NaOH, ref 4. [b] With 1.5 vol % dioxane.

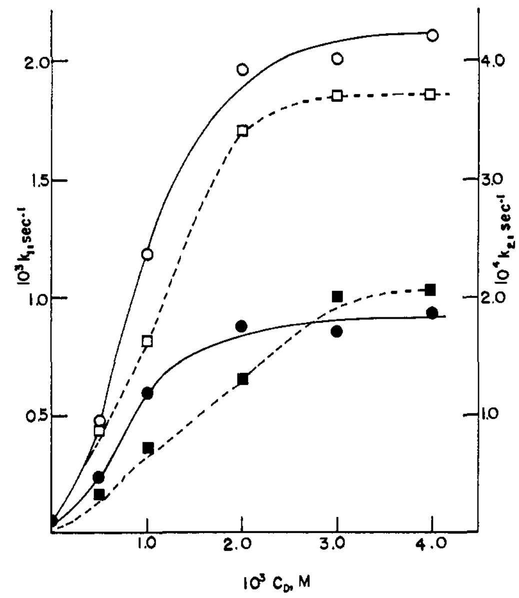

Figure 1. Values of k_1 (left-hand scale and solid line) and k_2 (right-hand scale and broken line) at 25.0° and 0.01 *M* NaOH. The open points refer to the solutions containing 1.5 wt % dioxane.

Table II. Micellar Catalysis of the Hydrolysis of the Reaction of 2,4-Dinitrophenyl Phosphate[a]

	10^3C_{CTABr}, *M*				
C_{OH^-}, *M*	0.50	1.00	2.00	3.00	4.00
		0.68	1.60	1.82	1.86
0.005			1.40		1.60
0.010	0.33	0.73	1.30	2.00	2.05
0.010[b]	0.88	1.62	3.40	3.70	3.70
0.020	0.33	0.83	2.40	2.80	2.50
0.100		1.50	2.27	2.90	2.40

[a] Values of 10^4k_2, sec^{-1} in aqueous solution at 25.0°. Values in the absence of OH$^-$ are from ref 7. [b] With 1.5 vol % dioxane.

We also measured the values of k_2 for the hydrolysis of 2,4-dinitrophenyl phosphate dianion at high concentrations of CTABr and CTACl up to 0.3 *M*, and

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(13) For a discussion of the use of this system see D. O. Harris, J. T. Gerig, and C. S. Ewig, *J. Chem. Educ.*, **47**, 97 (1970).

(14) C. A. Bunton and L. Robinson, *J. Amer. Chem. Soc.*, **90**, 5972 (1968).

(15) P. H. Elworthy, A. T. Florence, and C. B. MacFarlane, "Solubilization by Surface-Active Agents," Chapman and Hall, London, 1968, Chapter 1.

Table III. Effects of High Detergent Concentrations on the Hydrolysis of 2,4-Dinitrophenyl Phosphate[a]

	Detergent	
C_D, M	CTABr	CTACl
0.004	1.97	
0.005		1.88
0.01	1.90	
0.05	1.88	1.90
0.10	2.28	1.95
0.18	2.20	
0.40	2.26	

[a] Values of 10^4k_2, sec^{-1} at pH 9.0 in 0.015 M borate buffer.

found approximately constant values of k_2 for a given detergent (Table III). The values of k_2 at low detergent concentrations are slightly higher than those observed earlier,[6] probably because the borate buffer was 0.015 M instead of 0.025 M.

A few kinetic runs were made using 0.01 M sodium hydroxide in sodium lauryl sulfate, NaLS (Table IV).

Table IV. Effect of Anionic Detergent upon the Hydrolysis of Bis-2,4-dinitrophenyl and 2,4-Dinitrophenyl Phosphate[a]

C_{NaLS}, M	10^5k_1	10^6k_2
	3.0[b]	8.1[c]
0.005	2.6	6.1
0.010	2.3	5.0
0.025	2.9	6.4

[a] At 25.0° in 0.01 M NaOH. [b] Reference 4. [c] Reference 6.

This anionic detergent slightly reduced the values of k_1 and k_2. Similar observations were made earlier on the hydrolysis of the 2,6-dinitrophenyl phosphate dianion.[7] The kinetic effect of an uncharged detergent was also examined, using 0.01 M sodium hydroxide (Table V).

Table V. Effect of Uncharged Detergent upon the Hydrolysis of Bis-2,4-Dinitrophenyl Phosphate[a]

C_{DNPE}, M	10^5k_1
	3.0[b]
0.001	0.61
0.002	0.50
0.005	0.39
0.010	0.30
0.025	0.26

[a] At 25.0° in 0.01 M NaOH. [b] Reference 4.

Rather unexpectedly DNPE reduces the value of k_1, so that k_2 becomes larger than k_1, and the overall reaction follows approximate first-order kinetics. Under these conditions we cannot calculate good k_2 values from the kinetic data, although independent experiments showed that DNPE only slightly reduced the rate of the similar hydrolysis of 2,6-dinitrophenyl phosphate dianion.[7] The values of k_1 given in Table V were calculated using eq I, but they are very similar to the values obtained assuming a simple first-order kinetic form, and the first-order rate equation was followed for approximately two half-lives.

Added salts inhibit the catalysis of both steps of the hydrolysis by CTABr (Table VI), as is generally found.[7,11,12,14,16]

Table VI. Salt Effects upon Catalysis by CTABr[a]

	C_{salt}, M			
	0.02		0.04	
Salt	$k_1^{\mathrm{s}}/k_1^{\circ}$	$k_2^{\mathrm{s}}/k_2^{\circ}$	$k_1^{\mathrm{s}}/k_1^{\circ}$	$k_2^{\mathrm{s}}/k_2^{\circ}$
NaCl	0.75	0.93	0.43	0.71
NaBr	0.35	0.58	0.18	0.42
$NaNO_3$	0.27	0.40	0.14	0.24
NaOTos	0.05	0.15	0.011	0.045

[a] At 25.0° in aqueous solution with 0.01 M NaOH and 4 × 10^{-3} M CTABr.

In most of the experiments the nucleophile was hydroxide ion, but a few experiments were made at low pH where $k_2 \gg k_1$ and water is the nucleophile. Phosphate or borate buffers were used, and rate increases were observed (Table VII and VIII). The values of k_1 do not depend in any simple way upon the reagent or buffer concentrations, probably because phosphate ion may act as a nucleophile[11] (Scheme I), but it and borate ion should also hinder incorporation of bis-2,4-dinitrophenyl phosphate into the micellar phase.

Table VII. Effects of CTABr upon the Hydrolysis of Bis-2,4-dinitrophenyl Phosphate at pH 6.0[a]

10^3C_D, M	$10^5C_{\mathrm{substrate}}$, M	10^6k_1, sec^{-1}
0.05	0.6	1.5[b]
0.10	0.6	1.9[b]
0.25	0.6	2.7[b]
0.25	2.8	2.0[b]
0.50	2.8	1.9[b]
0.75	2.8	2.5[b]
1.0	2.8	2.5[b]
1.0	1.0	3.5[c]
2.0	1.0	3.2[c]
2.0	1.0	3.4[b]
5.0	1.0	2.0[c]
5.0	1.0	2.3[d]

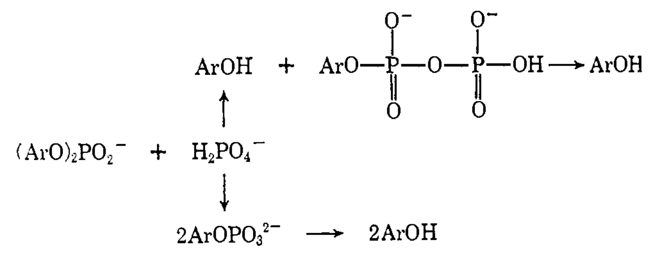
[a] In water at 25.0° with phosphate buffer; in the absence of detergent k_1 = 0.21 × 10^{-6} sec^{-1} at pH 6.0. [b] 1 × 10^{-3} M phosphate buffer. [c] 5 × 10^{-3} M phosphate buffer. [d] 2 × 10^{-4} M phosphate buffer.

Table VIII. Effects of CTABr upon the Hydrolysis of Bis-2,4-dinitrophenyl Phosphate at pH 8.0[a]

10^3C_D, M	$10^5C_{\mathrm{substrate}}$, M	10^6k_1, sec^{-1}
0.05	0.6	2.8
0.10	0.6	6.5
0.20	0.6	13
0.50	1.0	17
1.0	1.0	20
1.0	2.8	13

[a] In water at 25.0° with 2 × 10^{-3} M borate buffer; in the absence of detergent k_1 = 0.25 × 10^{-6} sec^{-1}.

Scheme I



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(16) C. A. Bunton and L. Robinson, *J. Org. Chem.*, 34, 780 (1969).

In addition a concurrent reaction involving C–O fission may occur, although other evidence makes this reaction improbable.[4] At pH 8.0 using borate buffer we observe large rate increases (Table VIII), probably because in the absence of detergent the major reaction involves attack of water upon the diaryl phosphate monanion,[4] whereas the cationic micelles so assist the reaction between hydroxide ion and the substrate that it becomes a major contributor to the overall reaction. These results show that hydrolysis at low pH is slower than at high pH, in the presence and absence of cationic micelles, although because of the complexity of the system we cannot separate the detergent effects upon the attack of water and hydroxide ion. For reactions in the aqueous phase the electrolyte effects of buffer ions can be compensated for, to a limited extent, by carrying out the reaction at constant ionic strength, or in the presence of a large concentration of electrolyte. This procedure is suspect because it ignores the specificity of many kinetic salt effects,[16,17] but it is completely unsatisfactory for micellar-catalyzed reactions where salts have large specific effects upon the incorporation of ionic reagents into the micellar phase.[7,11,12,16]

Turbidity Experiments. When bis-2,4-dinitrophenyl phosphate monoanion in low concentration is added to a slight excess of CTABr a precipitate is formed (Table IX), which does not coagulate on standing, and which exists as a stable suspension. The turbidity initially increases with added detergent, but then decreases at detergent concentrations above the critical micelle concentration (cmc), suggesting that the phosphate monoanion dissolves in the micellar phase giving a clear solution. These experiments were generally carried out with detergent concentrations considerably lower than those required to reach the plateau values of the rate constants.

Table IX. Turbidity Measurements on Mixtures of Bis-2,4-dinitrophenyl Phosphate and CTABr[a]

10^3C_{CTABr}, M	10^4C_s, M 1.09	1.65[b]	1.65[c]	1.65[d]	1.77	3.20	3.73[e]
0.000	0.000	0.000	0.000	0.000	0.00	0.00	0.03
0.196	0.012	0.028	0.040	0.020	0.10	0.13	
0.385	0.035	0.060	0.070	0.045	0.20	0.31	0.98
0.555					0.24		
0.566	0.050	0.075	0.075	0.055		0.40	
0.741	0.052	0.075	0.058	0.048	0.26	0.42	2.31
0.909	0.050	0.068	0.012	0.010	0.24	0.41	
1.07	0.042				0.22	0.39	1.83
1.23	0.029	0.055	0.000	0.000	0.18	0.36	
1.38	0.015				0.14	0.32	1.60
1.53	0.008				0.11	0.27	
1.66	0.000	0.030			0.04	0.08	1.04
1.81					0.01		
1.94					0.00		0.08
2.19							0.03
2.31		0.010					
2.86		0.000					

[a] Absorbance measured at 800 mμ unless specified. [b] 1 × 10^{-4} M NaOTos. [c] 5 × 10^{-4} M NaOTos. [d] 0.01 M NaCl. [e] Absorbance measured at 400 mμ.

The precipitate can form at detergent concentrations below the cmc and may be a sparingly soluble salt of phosphate monoanion and detergent cation, shown as an ion pair (A), with the following equilibria set up between the detergent cation, D^+, and the diaryl phosphate monoanion, P^-, and the substrate–micelle complex, PD_n^{n+}, but it is possible that a whole family of submicellar or other aggregates of substrate and detergent exist with no simple stoichiometry. Kinetic

$$D^+ + P^- \rightleftharpoons [D^+P^-] \rightleftharpoons [D^+P^-]\ \text{solid}$$

$$[D^+P^-] + (n-1)D^+ \rightleftharpoons P^+D_n^{n+}$$

and other evidence for such interactions have been obtained in other systems.[7,11,12,18,19] We found no turbidity when CTABr was mixed with dinitrophenyl phosphate dianions,[7] presumably because the hydration energy of these dianions is so high that they do not form ion pairs or aggregates analogous to A.

Because of the large rate inhibitions observed with added salts we also examined salt effects upon the turbidity of mixtures of CTABr and bis-2,4-dinitrophenyl phosphate. Added sodium tosylate sharply decreases the turbidity of solutions of CTABr and bis-2,4-dinitrophenyl phosphate, but sodium chloride has less effect, and sodium bromide and nitrate have intermediate effects. Some examples of this behavior are given in Tables IX–XIII for various concentrations of bis-2,4-dinitrophenyl phosphate, C_s, and added electrolytes. In addition we found that with 1.14 × 10^{-4} M bis-2,4-dinitrophenyl phosphate and CTABr concentrations up to 2.86 × 10^{-3} M the solutions remained clear over the whole range of detergent concentrations when 0.002 M or 0.01 M sodium tosylate was added.

Table X. Effect of Sodium Tosylate upon the Turbidity of Bis-2,4-dinitrophenyl Phosphate and CTABr[a]

10^4C_{NaOTos}, M	10^4C_s, M 0.91	1.95	2.65	5.53
0.00	0.050	0.095	0.31	0.98
0.98	0.042	0.092		
1.92	0.034	0.092		
2.83	0.020	0.089		
3.70	0.008	0.080	0.31	
4.55	0.000	0.068		
6.14		0.048		
7.63		0.028	0.30	
8.33		0.022		
11.3		0.004	0.27	
14.8			0.23	
21.4			0.15	
27.6			0.080	
33.3			0.024	
63.7				0.75
235				0.56
333				0.41
500				0.27
636				0.165

[a] Values of absorbance measured at 800 mμ in the presence of 8 × 10^{-4} M CTABr. Alternate points are omitted for the experiments at the higher concentrations of sodium tosylate.

The results in Tables IX–XIII show that the amount of sodium salt which is needed to remove the turbidity increases sharply with increasing concentration of bis-2,4-dinitrophenyl phosphate, probably because high

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(17) C. A. Bunton, J. H. Crabtree, and L. Robinson, *J. Amer. Chem. Soc.*, **90**, 1258 (1968); C. A. Bunton and L. Robinson, *ibid.*, **90**, 5165 (1968), and references cited.

(18) P. Mukerjee and K. J. Mysels, *ibid.*, 77, 2937 (1955).

(19) T. C. Bruice, J. Katzhendler, and L. R. Fedor, *ibid.*, **90**, 133 (1968).

Table XI. Effect of Sodium Chloride upon the Turbidity of Bis-2,4-dinitrophenyl Phosphate and CTABr[a]

10^2C_{NaCl}, M		10^4C_s, M	
	1.65	2.90	4.84
0.00	0.075	0.200	0.46
0.49	0.070		
0.96	0.050	0.200	
1.42	0.025		
1.92	0.000	0.180	0.44
2.83		1.555	
3.85		0.127	0.45
7.63		0.045	0.75
8.33		0.015	
9.02		0.005	0.80
10.7		0.000	0.75
13.8			0.67
16.2			0.58
19.4			0.49
21.9			0.42
26.5			0.31

[a] Values of absorbance measured at 800 mμ in the presence of 8×10^{-4} M CTABr. Alternate points are omitted for the experiments at higher concentrations of NaCl.

Table XII. Effect of Sodium Nitrate upon the Turbidity of Bis-2,4-dinitrophenyl Phosphate and CTABr[a]

$10^3C_{NaNO_3}$, M	10^4C_s, M	
	1.40	2.80
0.00	0.075	0.200
0.99	0.075	
2.91	0.070	
4.77	0.045	
6.54	0.022	
8.26	0.008	
9.91	0.000	
14.8		0.190
23.6		0.112
28.0		0.085
32.2		0.042
36.3		0.015
40.3		0.000

[a] Values of absorbance measured at 800 mμ in the presence of 8×10^{-4} M CTABr.

Table XIII. Effect of Sodium Bromide upon the Turbidity of Bis-2,4-dinitrophenyl Phosphate and CTABr[a]

10^3C_{NaBr}, M	10^4C_s, M	
	1.40	2.80
0.00	0.070	0.200
1.96	0.068	
2.91	0.065	
3.84	0.062	
5.66	0.048	0.200
7.42	0.028	
9.80	0.011	0.190
14.6	0.000	0.162
19.2		0.128
28.3		0.075
37.1		0.200
45.4		0.000

[a] Values of absorbance measured at 800 mμ in the presence of 8×10^{-4} M CTABr. Alternate points are omitted for higher concentrations of NaBr.

concentrations of the bisphosphate will stabilize these coacervates or other aggregates between the detergent and phosphate,[20] relative to the micellar phase. Independent experiments showed that solutions of CTABr and sodium chloride or tosylate are clear over the range of concentrations used, but it is possible that there may be coprecipitation of chloride or tosylate ion with bis-2,4-dinitrophenyl phosphate salts of the cetyltrimethylammonium ion. However the simpler explanation of these results is that a large anion, such as tosylate, interacts sufficiently strongly with monoanionic or micellized CTABr to prevent it from forming insoluble salts with the substrate, whereas the smaller chloride ion is much less effective. The added salt should promote micellization, and the diaryl phosphate could be taken up into these micelles. However this effect should not be too important in reducing turbidity, because added salts actually exclude anionic phosphate esters from the micellar phase (Table VI and ref 7).

With relatively high concentrations of CTABr (8×10^{-4} M) and phosphate ester (4.84×10^{-4} M), added sodium chloride at first increases the turbidity, probably by "salting out" A, and the turbidity is not removed even by 0.26 M sodium chloride. These results suggest that we are not dealing with a simple ionic strength effect, but that bromide, nitrate, and especially tosylate are much more effective than chloride ions in interacting with CTA^+ and we note that rate inhibition by anions shows that interaction between anion and the detergent increases in the order of $Cl^- < NO_3^- \approx Br^- < OTos^-$ (Table VI and ref 7,11, 14,16).

The salt effects upon turbidity of mixtures of CTABr and bis-2,4-dinitrophenyl phosphate are similar to those on the rates of CTABr-catalyzed reactions, because both effects depend upon interactions between the cationic micelle and the added counteranion. However information is needed about the composition of the coacervates before the salt effects can be explained completely, and these salt effects on turbidity can be used alongside salt inhibition studies as probes for studying the interactions between anions and cationic detergents.

Discussion

In alkaline solution we were able to follow both steps of the hydrolysis of bis-2,4-dinitrophenyl phosphate to inorganic phosphate. However, the results for the second step, the hydrolysis of 2,4-dinitrophenyl phosphate dianion, for the most part confirm those obtained earlier (Table II and ref 7), and the discussion is therefore focused largely on the first step of the reaction.

Reactions of Bis-2,4-dinitrophenyl Phosphate. The catalysis of the reaction of bis-2,4-dinitrophenyl phosphate with hydroxide ion by micelles of CTABr can be explained in terms of the ability of the cationic micelle to bring the anionic reagents together in an environment in which the transition state will be stabilized relative to the initial state. The transition state is a dinegatively charged ion whose charge density should be lower than those of the reactants, and therefore the free energy of activation should be lower in the micellar phase than in water. The simple theory of micellar catalysis assumes that reaction takes place either in the aqueous phase or in the micellar pseudophase, and that there should be neither catalysis nor inhibition at detergent concentrations below the cmc, where only detergent monomers are assumed to be present. There is considerable qualitative, and some quantitative, support for this simple treatment.[7,11,12,14,16,21-23]

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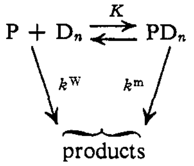
[20] Reference 15, p 209.

It is assumed that the concentration of micelles, C_m, is given by eq II, and provided that the phosphate monoanion, P, associates only with micelles (Scheme II)

$$C_m = (C_D - \text{cmc})/N \tag{II}$$

we can relate k_1 and C_m using eq III where k^W and k^m are the rate constants for reaction in the aqueous and micellar phases, respectively.

Scheme II



$$k_1 = \frac{k^W + k^mKC_m}{1 + KC_m} \tag{III}$$

In the plateau region where $C_D > 0.002$ *M* and $KC_m \gg 1$ and $k^mKC_m \gg k^W$ the first-order rate constant, k_1, for reaction of bis-2,4-dinitrophenyl phosphate is approximately proportional to hydroxide ion concentration at $C_{OH^-} < 0.02$ *M*, but at higher hydroxide ion concentrations there is little increase in k_1 (Table I). These results are readily understandable because high concentrations of hydroxide ion can saturate the cationic micelles,[14] and an anion, such as hydroxide, will also inhibit the incorporation of the diaryl phosphate monoanion into the micellar phase. Therefore, hydroxide ion acts both as the reagent and as an inhibitor, although because of its high charge density it should not be a very effective inhibitor.[11] In the plateau region k_1 is increased approximately 30-fold by the addition of CTABr. This rate enhancement is similar to those (10–10^2) generally found for nucleophilic attack by small anions[12,14,16,21–23] (*cf.* ref 24, 25).

Although rate maxima are generally found in reactions catalyzed by ionic micelles we found none in this reaction, probably because the plateau values of k_1 are reached at much lower detergent concentrations than those generally needed.[12,14,16,23] This question is discussed later in this paper.

Interactions between the Substrate and CTABr. The simple theory which leads to eq II and III is not quantitatively correct in the present system, because there is extensive catalysis by CTABr below the cmc (Table I). (For CTABr in water, cmc = 7.8×10^{-4} *M* and 3.2×10^{-4} *M* in 0.05 *M* NaOH.)[14] These rate enhancements could be caused by (i) a lowering of the cmc by the substrate or (ii) the formation of submicellar aggregates. There is very strong evidence for the formation of submicellar aggregates in other systems[18] and many workers have noted that they could be catalytically important,[11,12,19] and that they could be responsible for the sigmoid form of plots of rate constant *vs.* detergent concentration at low detergent concentrations. The turbidity experiments (Tables IX–XIII) suggest that submicellar aggregates are important in the present system, and that they are sensitive to the electrolytes present.

Because of their higher hydration energy the dinitrophenyl phosphate dianions should be less effective than the bis-2,4-dinitrophenyl phosphate monoanion in associating with monomeric CTABr, or promoting its micellization, and their hydrolyses are only slightly catalyzed by CTABr concentrations below the cmc,[7] and therefore the simple treatment is more effective for their hydrolyses.

Effects of Anionic and Unchanged Detergents. The small kinetic effect of NaLS is readily understandable because both hydroxide and bis-2,4-dinitrophenyl phosphate ions should be repelled by the anionic micelles, and reaction should take place only in the aqueous phase. The results also suggest that detergents do not affect reaction rates indirectly, for example by changing the nature of the solvent. These conclusions agree with existing evidence.[12,14,16,21–23]

The rate retardations by the uncharged micelles (Table V) were unexpected. The hydrolysis of 2,6-dinitrophenyl phosphate dianions is retarded only slightly by DNPE,[7] and even though uncharged substrates may be taken up into the micellar phase when uncharged detergents are used, the reaction rate is rarely affected.[14,22,23] Exceptions to this generalization are the reactions of *p*-nitrophenyldiphenyl phosphate with hydroxide and fluoride ions,[11] which are strongly retarded by uncharged micelles of DNPE, and the reaction of 1,1,1-trichloro-2-methyl-2-propanol with hydroxide ion in the presence of a polyether,[26] probably because the sparingly soluble substrate is taken up into the interior of the micelles where it is protected from an anionic nucleophile. It seems that the same thing is happening with bis-2,4-dinitrophenyl phosphate, despite its negative charge, and if this explanation is correct it means that interactions between an uncharged micelle and bulky organic residues can be strong enough to overcome hydration of the negative charge, and there is evidence for the solubilization of low charge density amine cations and aryl sulfate ions by uncharged polysorbate micelles.[27] We note here that the inhibition by DNPE is greater for the uncharged *p*-nitrophenyldiphenyl phosphate than for the anionic bis-2,4-dinitrophenyl phosphate.[11]

These results show that the observations of micellar catalysis or inhibition depend not only upon the reactants being taken up by the detergent, but also upon the region of micelle which they occupy, and the extent to which the micelle can stabilize or destabilize the transition state. There are many examples of reactions in which incorporation of reactants into the micellar phase has little, or no, kinetic consequence.[14,16,22,23,28] In many of these cases the substrate is probably in the outer, water-rich region of the micelle, where it could be exposed to the reagent,[29] but in some special cases the substrate is taken up into a

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[21] E. F. Duynstee and E. Grunwald, *J. Amer. Chem. Soc.*, **81**, 4540, 4542 (1959).

[22] D. G. Herries, W. Bishop, and F. M. Richards, *J. Phys. Chem.*, **68**, 1842 (1964).

[23] F. M. Menger and C. E. Portnoy, *J. Amer. Chem. Soc.*, **89**, 4698 (1967).

[24] C. Gitler and A. Ochoa-Solano, *ibid.*, **90**, 5004 (1968).

[25] We note here that the calculated magnitude of the (very large) micellar catalysis observed by Gitler and Ochoa-Solano[24] depends on assumptions made as to the nature of the micelle–substrate complex.

[26] R. A. Anderson and A. M. Slade, *J. Pharm. Pharmacol.*, **18**, 640 (1966).

[27] Reference 15, p 179.

[28] E. J. Fendler, J. H. Fendler, and R. R. Liechti, *J. Org. Chem.*, **35**, 1658 (1970).

[29] J. C. Erickson and G. Gilberg, *Acta Chem. Scand.*, **20**, 2019 (1966); M. Donborne and C. T. Rhodes, *J. Chem. Soc.*, 6166 (1964); *J. Pharm. Pharmacol.*, **18**, 424 (1966); A. S. Waggoner, O. M. Griffith, and C. R. Christiansen, *Proc. Natl. Acad. Sci.*, **57**, 1198 (1967).

region of an uncharged micelle in which it is protected from an anionic nucleophile. Alternatively the substrate may stay in the outer region of the micelle, but be oriented in such a way that it does not react with an external reagent. Spectroscopic rather than kinetic methods are needed to differentiate between these two possibilities.

Kinetic Salt Effects. Added salts reduced the extent of micellar catalysis by CTABr (Table VI), as is generally observed, and their effect increases with decreasing charge density of the counterion.[7,11,12,14,16] Catalysis of the reaction between hydroxide and the bis-2,4-dinitrophenyl phosphate ions requires that both anions come together on the cationic micelle. Because one ion hinders the attachment to the micelle of another ion of like charge each of the reagents would inhibit attachment of the other, and added counteranions should have an especially large inhibiting effect, because they will hinder the attachment of both reagents. Consistently, salt effects are much larger for the reaction of hydroxide ion with the diaryl phosphate monoanion than for the spontaneous hydrolysis of 2,4-dinitrophenyl phosphate dianion, as is shown by the smaller values of k_1^s/k_1° as compared with k_2^s/k_2° for a given salt (Table VI). The values of k_2^s/k_2° for the hydrolysis of the 2,4-dinitrophenyl phosphate dianion determined in this work are consistent with those observed earlier.[7]

Salt inhibition increases in the order NaCl $<$ NaBr $\sim$ $NaNO_3$ $<$ NaOTos, which is also the order in which these salts hinder the formation of submicellar aggregates of CTA and bis-2,4-dinitrophenyl phosphate (Tables IX–XIII). Both phenomena depend upon the increasing interaction of an anion with a cation or a cationic micelle with decreasing charge density of the anion.

Hydrolysis of the 2,4-Dinitrophenyl Phosphate Dianion. The simple kinetic treatment for micellar catalysis, based on eq II and III, ignores changes in micellar shape or aggregation number with increasing concentration of an ionic detergent. For many detergent-catalyzed reactions between an unchanged substrate and an ion, rates reach maxima at detergent concentrations in the region 0.01–0.1 *M*,[12,14,16,23] either because of effects due to counterions,[12] or to deactivation of the ionic reagent,[14] but the effects could also be related to changes in micellar structure.[15] The CTABr-catalyzed hydrolysis of 2,4-dinitrophenyl phosphate dianion is a good probe for the study of kinetic effects caused by changes in micellar structure. Micelles of CTABr are spherical at low concentrations, but they become rodlike at 5% w/w, *i.e.*, at *ca.* 0.14 *M*,[15] and therefore we might expect to find large rate changes here, because, for example, the surface area to volume ratio of the micelle should change. However, the values of k_2 do not change sharply at high detergent concentrations (Table III), although k_2 increases by *ca.* 20% at a CTABr concentration of 0.1 *M*, and then levels off, possibly because of the structural change in the micelle at this concentration. In addition the relatively high concentration of bromide ions in 0.1–0.3 *M* CTABr has no obvious kinetic effect, suggesting that the important factor in anion inhibition is not the actual anion concentration, but that relative to the concentration of detergent cations, as can be seen by comparing the kinetic data in Tables III and VI. For these reasons we believe that the rate maxima found for detergent-catalyzed ion–molecule reactions are not caused by increasing concentration of counterion, but by inhibition of the ionic reagent,[14] and that the kinetic relations are not very sensitive to changes in micellar shape brought about by changes in detergent concentration.

The rate measurements using cetyltrimethylammonium chloride, CTACl (Table III), show that a change of counteranion from bromide to chloride has almost no effect, although CTACl is a somewhat better catalyst than CTABr in nucleophilic aromatic substitution.[30] We do not know whether the shape of CTACl micelles changes at high concentration,[15] but if it does the effects upon catalysis are small.

A problem in attempting to use the simple treatments based on eq II and III to explain micellar catalysis and inhibition is that possible changes in micellar size or structure are ignored. The present results suggest that the simple assumptions are reasonably satisfactory.

Reaction of Hydroxide Ion with 2,4-Dinitrophenyl Phosphate. In the absence of detergents, attack of hydroxide upon a dinitrophenyl phosphate dianion makes only a small contribution to the overall reaction, except at high concentrations of hydroxide ion.[6] With 2,6-dinitrophenyl phosphate hydroxide ion does not increase the rate of reaction in the presence of CTABr,[7] but there is some rate enhancement with 2,4-dinitrophenyl phosphate (Table II). (The major part of the increase in rate constant from 8×10^{-6} sec^{-1}, for the spontaneous hydrolysis of 2,4-dinitrophenyl phosphate dianion in the absence of detergent,[6] to the values given in Table II, is, however, caused by detergent catalysis of the spontaneous hydrolysis of the dianion.[7]) The overall rate effects are small because although hydroxide ion can attack the phosphate dianion in the micellar phase it inhibits incorporation of the dianion into the micellar phase, and therefore tends to suppress the kinetically more significant spontaneous hydrolysis of the dianion, so that its overall rate effect is the sum of two opposing effects.

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(30) C. A. Bunton and L. Robinson, *J. Amer. Chem. Soc.*, **91**, 6072 (1969).