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CRDEC-TR-096

THE CATALYTIC HYDROLYSIS OF p-NITROPHENYLDIPHENYL PHOSPHATE IN A CTAB MICROEMULSION

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June 1989

U.S. ARMY
ARMAMENT
MUNITIONS
CHEMICAL COMMAND

Aberdeen Proving Ground, Maryland 21010-5423

UNCLASSIFIED
SECURITY CLASSIFICATION OF THIS PAGE

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1a. REPORT SECURITY CLASSIFICATION
UNCLASSIFIED

1b. RESTRICTIVE MARKINGS

2a. SECURITY CLASSIFICATION AUTHORITY

2b. DECLASSIFICATION / DOWNGRADING SCHEDULE

3. DISTRIBUTION / AVAILABILITY OF REPORT
Approved for public release; distribution is unlimited.

4. PERFORMING ORGANIZATION REPORT NUMBER(S)
CRDEC-TR-096

5. MONITORING ORGANIZATION REPORT NUMBER(S)

6a. NAME OF PERFORMING ORGANIZATION
CRDEC

6b. OFFICE SYMBOL *(If applicable)*
SMCCR-RS

7a. NAME OF MONITORING ORGANIZATION

6c. ADDRESS *(City, State, and ZIP Code)*
Aberdeen Proving Ground, Maryland 21010-5423

7b. ADDRESS *(City, State, and ZIP Code)*

8a. NAME OF FUNDING / SPONSORING ORGANIZATION
CRDEC

8b. OFFICE SYMBOL *(If applicable)*
SMCCR-RS

9. PROCUREMENT INSTRUMENT IDENTIFICATION NUMBER

8c. ADDRESS *(City, State, and ZIP Code)*
Aberdeen Proving Ground, Maryland 21010-5423

10. SOURCE OF FUNDING NUMBERS

PROGRAM ELEMENT NO.	PROJECT NO.	TASK NO.	WORK UNIT ACCESSION NO.
	1C162706	A553F	

11. TITLE *(Include Security Classification)*
The Catalytic Hydrolysis of p-Nitrophenyldiphenyl Phosphate in a CTAB Microemulsion

12. PERSONAL AUTHOR(S) Mackay, Raymond A., Ph.D.; Durst, H. Dupont, Ph.D. (Research Directorate); Longo, Frederick R., Ph.D. (Drexel University); (continued on reverse)

13a. TYPE OF REPORT
Technical

13b. TIME COVERED
FROM 86 Sep TO 87 Oct

14. DATE OF REPORT *(Year, Month, Day)*
1989 June

15. PAGE COUNT
18

16. SUPPLEMENTARY NOTATION

17. COSATI CODES

FIELD	GROUP	SUB-GROUP
07	02	
20	06	

18. SUBJECT TERMS *(Continue on reverse if necessary and identify by block number)*

Hydrolysis	Kinetics	Iodosobenzoic acid
Microemulsions	Catalysis	(continued on
Micellar solutions	Phosphate esters	reverse)

19. ABSTRACT *(Continue on reverse if necessary and identify by block number)*

The iodosobenzoate catalysis of the hydrolysis of p-nitrophenyldiphenyl phosphate in an oil-in-water microemulsion stabilized by cetyltrimethylammonium bromide and 1-butanol has been examined. In addition, we have investigated this hydrolysis in water and in cetyltrimethylammonium chloride micellar solutions. The second-order rate constants for the catalytic and non-catalytic pathways have been determined. Direct tests demonstrate that iodosobenzoate is a turnover catalyst.

20. DISTRIBUTION / AVAILABILITY OF ABSTRACT
☒ UNCLASSIFIED/UNLIMITED ☐ SAME AS RPT. ☐ DTIC USERS

21. ABSTRACT SECURITY CLASSIFICATION
UNCLASSIFIED

22a. NAME OF RESPONSIBLE INDIVIDUAL
SANDRA J. JOHNSON

22b. TELEPHONE *(Include Area Code)*
(301) 671-2914

22c. OFFICE SYMBOL
SMCCR-SPS-T

DD Form 1473, JUN 86 *Previous editions are obsolete.*

SECURITY CLASSIFICATION OF THIS PAGE
UNCLASSIFIED

12. PERSONAL AUTHOR(S) (continued)

and Knier, Barry L. (Universtiy of Puerto Rico)

18. SUBJECT TERMS (continued)

Phosphate esters
Iodosobenzoic acid
Catalytic pathways
Second-order rate constants
Decontamination
Turnover catalyst
Non-catalytic pathways
Chemical warfare

PREFACE

The work described in this report was authorized under Project Number 1C162706A553F, CB Decontamination and Contamination Avoidance. This work was started in September 1986 and completed in October 1987. The experimental data are recorded in laboratory notebook 85-0173.

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The Catalytic Hydrolysis of p-Nitrophenyldiphenyl Phosphate in a CTAB Microemulsion

1 INTRODUCTION

During the past decade, mechanistic studies of several phosphate and phosphonate esters have been reported. These substances are known to be acetylcholine esterase inhibitors and are useful in mechanistic studies of enzyme action.[1] Additionally, their related neurotoxicity makes them important as pesticides,[2] and their decomposition is similarly important. Since these phosphates are normally only slightly soluble in water, micellar and microemulsion media have often been employed in mechanistic investigations.[3,4] Under these conditions enhancements are observed in both solubilization and in the rate of decomposition. This micellar catalysis is thought to involve a local increase in the concentrations of the reactive species at the micellar surface.

One particularly interesting adaptation of this micellar approach is the use of 2-iodosobenzoic acid (IBA) and its derivatives as nucleophilic catalysts in phosphate ester decomposition. The work by Moss and co-workers[5] employed both IBA, its 5-octoxy- and surfactant functionalized derivatives in an aqueous cetyltrimethylammonium chloride (CTAC) micellar medium. Their data strongly indicate that, at least in a micellar medium, IBA accelerates the rate of ester decomposition without being consumed.

The peculiarities in the structure of IBA undoubtedly contribute to its catalytic activity. X-ray studies[6,7] indicate that as the crystalline sodium salt it exists in the bicyclic form **1b** (Scheme I of Figure 1). In solution the pK_a of IBA is 7.02 (in our hands, by titrimetry). This is a strong indication that the "normal" IBA structure (**1a**) is unfavorable in solution as well since (1) the upper limit for pK_a values for substituted benzoic acids is about 5.5, and (2) the iodoso substituent should be electron-withdrawing and thus monocyclic IBA, **1a**, should have a pK_a less than 4.2, the pK_a of unsubstituted benzoic acid.

The accepted mechanism for the uncatalyzed hydrolysis of PNDP, although it has not been definitively proven, involves attack of nuclophile X^- on PNDP, accompanied or followed by expulsion of the 4-nitrophenoxide ion,[8,9] illustrated in Scheme II of Figure 1. In the presence of catalyst IBA the mechanism is believed to involve attack of IBA on phosphorus followed by release of 4-nitrophenoxide. Regeneration of the catalyst involves nucleophilic attack on the IBA-phosphate intermediate, as shown in Scheme III (Figure 1).

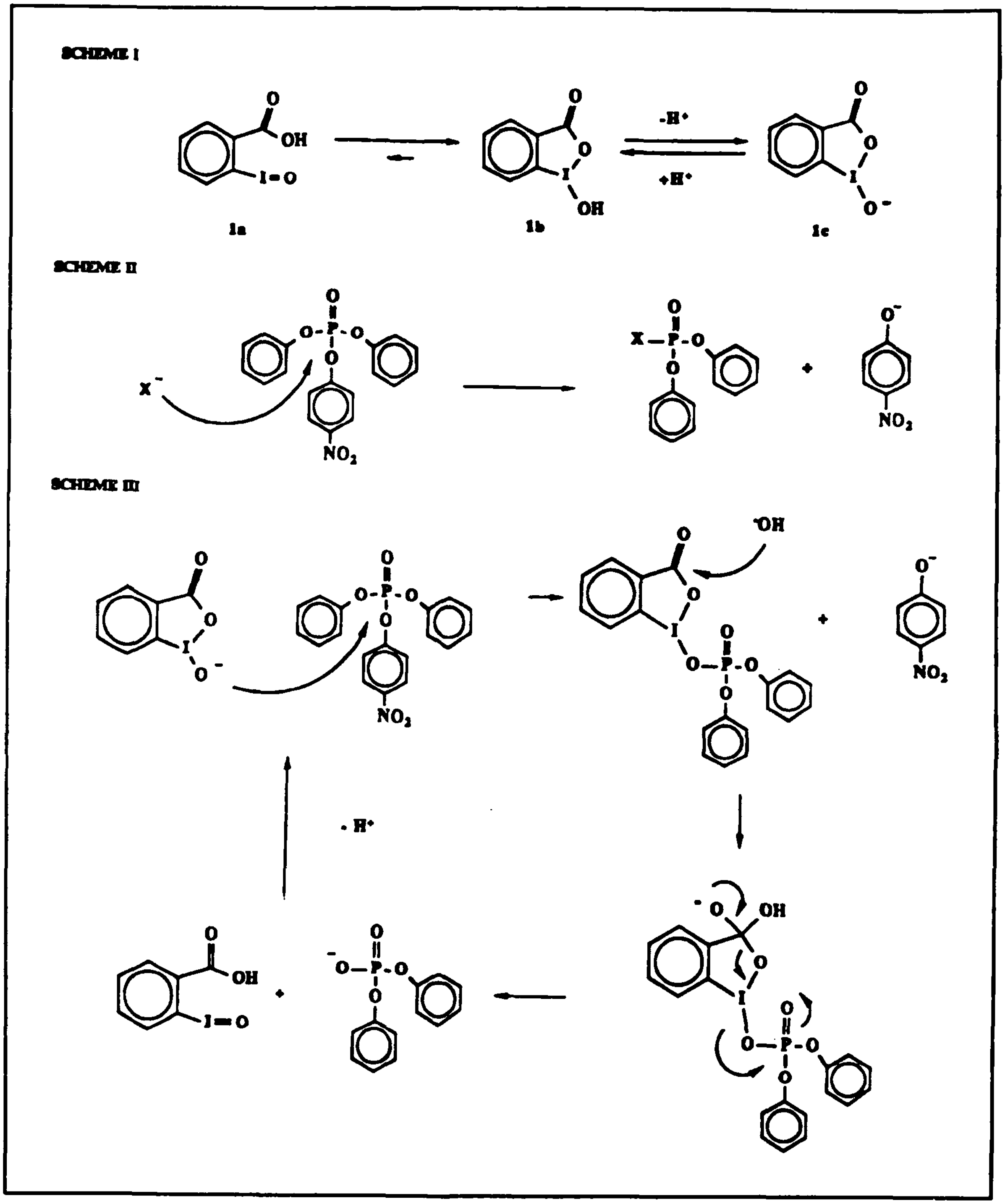

Figure 1. Schemes Demonstrating Chemical Transformations

We have extended this work to microemulsion media by examining the decomposition of 4-nitrophenyldiphenyl phosphate (PNDP) in a hexadecane/water microemulsion stabilized by cetyltrimethylammonium bromide (CTAB) and 1-butanol. This medium was chosen because it was previously used in the absence of IBA[8] and thus serves as a base line for the catalytic investigations.

2 EXPERIMENTAL SECTION

Cetyltrimethylammonium bromide (CTAB) was purchased from Aldrich. Microemulsions prepared from this material as received gave kinetic data indistinguishable from microemulsions which had been prepared from CTAB recrystallized from methanol. The 1-butanol and *n*-hexadecane were Aldrich reagent grade and were used as received. The iodosobenzoic acid (IBA) was obtained from Sigma and used as supplied. The aqueous phase was 0.03 M borate buffer prepared by using sodium tetraborate decahydrate ($Na_2B_4O_7 \cdot 10H_2O$, Baker Reagent) and deionized, glass-distilled water. In most of the experiments a microemulsion consisting of 18% (by mass) CTAB, 18% 1-butanol (cosurfactant), 4% *n*-hexadecane (oil), and 60% aqueous buffer was used. The pH values reported are those determined directly in the microemulsion, the pH of which was generally 0.2 unit higher than the aqueous buffer from which it was prepared.[10] The initial concentration of PNDP was normally 5.44×10^{-5} M. Experiments designed to give the hydroxide rate constant employed microemulsions prepared (and titrated) in the range of 1.2×10^{-3} to 1×10^{-2} M in NaOH.

Reactions were followed by monitoring the absorbance of the product, 4-nitrophenoxide ion, at 402 nm with either a Hewlett-Packard 8450A photodiode array spectrophotometer or a Gilford 2400S spectrophotometer equipped with a thermostated four-cell carriage and cell positioning programmer. The Gilford 2400S spectrophotometer was interfaced to an Apple IIe computer via an Adalab Interface card for "real time" data acquisition. Temperature control (25 ± 0.1 °C) was achieved by use of an Endocal RTE-8DD controlled bath.

Rate constants were generated on an Apple IIe microcomputer using a weighted linear least-squares technique applied to the data in the $\ln (A_{inf} - A_t)$ vs time form. Individual weights were calculated by using a linear-to-log transform procedure.[11]

The pH measurements were performed by using a glass electrode with an Orion or Fisher pH meter, calibrated with standard buffers at pH values of 4.00, 7.00, and 10.00, also obtained from Fisher.

3 RESULTS AND DISCUSSION

The decomposition of PNDP in aqueous or largely aqueous media can be reasonably expected to follow a rate law of the form:

$$\text{rate} = k_{hyd}\,[\text{PNDP}] + k_{OH}\,[\text{OH}^-]\,[\text{PNDP}] + k_{IBA}\,[\text{IBA}]\,[\text{PNDP}]$$

$$= \{\, k_{hyd} + k_{OH}\,[\text{OH}^-] + k_{IBA}\,[\text{IBA}]\,\}\,[\text{PNDP}] \qquad (1)$$

where the term $k_{hyd}[\text{PNDP}]$ represents the hydroxide independent "background" reaction, the $k_{OH}[\text{OH}^-][\text{PNDP}]$ term represents the second-order reaction between OH^- and PNDP, and the $k_{IBA}[\text{IBA}][\text{PNDP}]$ term represents the rate enhancement resulting from the addition of IBA to the system. By appropriately choosing the experimental conditions, each of these microscopic rate constants were independently determined.

In order to obtain the k_{IBA} value, experiments were run at several buffered pH values. Under these conditions the first two terms of equation 1, $k_{hyd} + k_{OH}[\text{OH}]$, are constant. If, as Moss *et al.* suggest,[12] IBA is not consumed in the reaction, then the third term is also constant, and Equation 1 reduces to a description of an experimentally first-order process:

$$\text{rate} = k_{obsd}[\text{PNDP}] \qquad (2)$$

where

$$k_{obsd} = k_{hyd} + k_{OH}\,[\text{OH}^-] + k_{IBA}\,[\text{IBA}] \qquad (3)$$

Figure 2 shows plots of absorbance-time data at several different IBA concentrations in the microemulsion system. The lines drawn through the points are theoretical lines assuming simple first-order kinetics as in equation 2. Figure 3 shows the same data plotted as log $(A_{inf} - A_t)$ vs. time. Such linearity was normally observed for five or more half-lives indicating no significant deviation from first-order behavior. This is evidence that IBA is a "turn-over" catalyst.

A plot of k_{obsd} vs. [IBA] at pH 8.5 yields a straight line (Figure 4) as predicted by equation 3 having a slope equal to k_{IBA} and an intercept equal to $k_{hyd} + k_{OH}[\text{OH}^-]$.

To ensure that the preliminary analysis was valid, we collected similar data at pH values of 9.2, 9.8, and 10.5. The value of k_{IBA} should be independent of pH since the pK_a of IBA is 7.02 and, therefore, is fully ionized at each of the pHs under study.

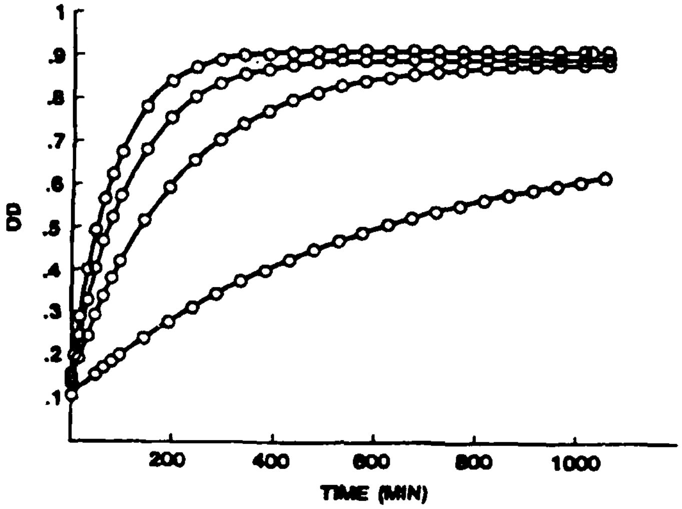

Figure 2. A Plot of Absorbance of 4-Nitrophenoxide vs Time at pH 9.2 and at Varying Values of [IBA]; the Solid Lines are Calculated on the Assumption of First-Order Behavior

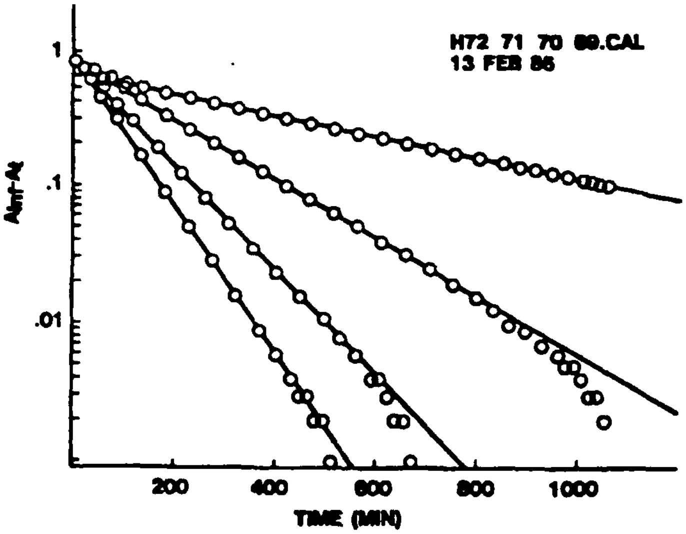

Figure 3. A Plot of the Log (A_{inf} - A_t) vs Time Obtained by Using the Data of Figure 2. Note that First-Order Behavior is Exhibited for Several Half-Lives.

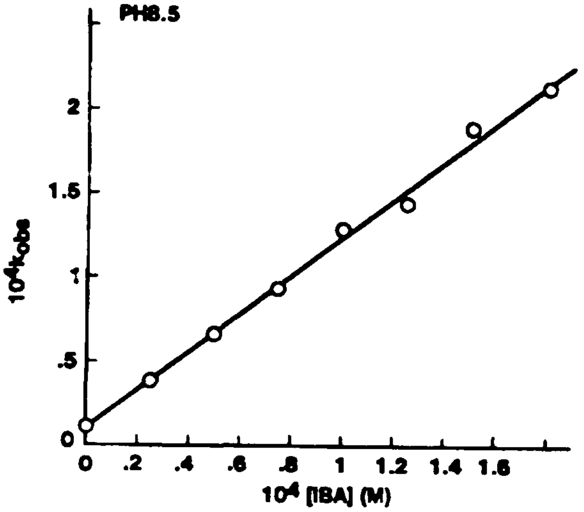

Figure 4. The Plot of k_{obsd} Vs [IBA] (Linear at pH 8.5 and at All pHs Examined). The Slope of the Line Is Equal to k_{IBA}; the Intercept Equals k_{hyd} + k_{OH} [OH^-].

The values of k_{IBA} and (k_{hyd} + k_{OH}·[OH^-]), as determined from analyses of at least eight experiments at each pH, are presented in Table 1. Since the errors in the slopes of the plots of k_{obsd} vs. [IBA] are in the range of 2 to 5%, these data present no evidence of any variation in k_{IBA} in the pH range 8.5 to 10.5. The average value of the slopes (and the average value of k_{IBA}) is 1.22 ± 0.07 $M^{-1}s^{-1}$.

In order to get more reliable values for k_{hyd} and k_{OH}, microemulsions were prepared with standard NaOH solution containing no catalyst as the aqueous phase. Kinetic experiments were performed with [OH^-] varying from 1.20×10^{-3} to 1.00×10^{-2} M. Since there is no IBA present, equation 1 becomes:

$$\text{rate} = k_{hyd}[\text{PNDP}] + k_{OH}[\text{OH}^-][\text{PNDP}] \qquad (4)$$

or

$$\text{rate} = k_{obsd}[\text{PNDP}]$$

where

$$k_{obsd} = k_{hyd} + k_{OH}[\text{OH}^-] \qquad (5)$$

Table 1. Effect of pH on k_{OH} and k_{IBA} in CTAB Microemulsion at 25 °C

pH	$k_{hyd} + k_{OH}$ [OH-] X 10^5 / s^{-1}	k_{IBA} / s^{-1} M^{-1}
8.5	0.93 ± 0.28	1.14 ± 0.03
9.2	2.52 ± 0.22	1.31 ± 0.03
9.8	4.05 ± 0.51	1.24 ± 0.05
10.5	4.84 ± 0.51	1.18 ± 0.03

Since we used a high excess of $[OH^-]$ in these experiments, the rate law was pseudo first order. Figure 5 depicts the dependence of k_{obsd} on $[OH^-]$. The least-squares slope of this line is equal to k_{OH} with a best fit value of 0.145 $M^{-1}s^{-1}$, which is in good agreement with the value extracted from previously published data.[8,13] The intercept, k_{hyd}, is 5 X 10^{-6}, which is indistinguishable from zero for these data. Hence, equation 1 may be written as:

$$\text{rate} = \{0.145\,[OH-] + 1.22\,[IBA]\}\,[PNDP] \qquad (6)$$

Analogous experiments were performed in aqueous solution for comparison. The results from studies of buffered solutions (again for the determination of k_{IBA}) are listed in Table 2, and the results from studies of NaOH solutions (for the determination of k_{hyd} and k_{OH}) are in Table 3. The value of k_{IBA} in aqueous solution is smaller than in the microemulsion and has a slight, but real, dependence on pH while k_{OH} is 0.437 $M^{-1}s^{-1}$.

The initial work by Moss et al.[5,11] gave observed rate constants as a function of [CTAC]. We chose to expand this work to measure second-order rate constants for direct comparison. Four different micellar CTAC solutions were employed and kinetic experiments were performed at four IBA concentrations in each of these media, thus allowing the determination of both k_{IBA} and the combined $k_{hyd} + k_{OH}$ $[OH^-]$ terms at each CTAC concentration. All runs were buffered with 0.03 M borate at pH 9.2. The results summarized in Table 4 show that k_{IBA} decreases with increasing [CTAC], which is the expected result above the critical micelle concentration (cmc).

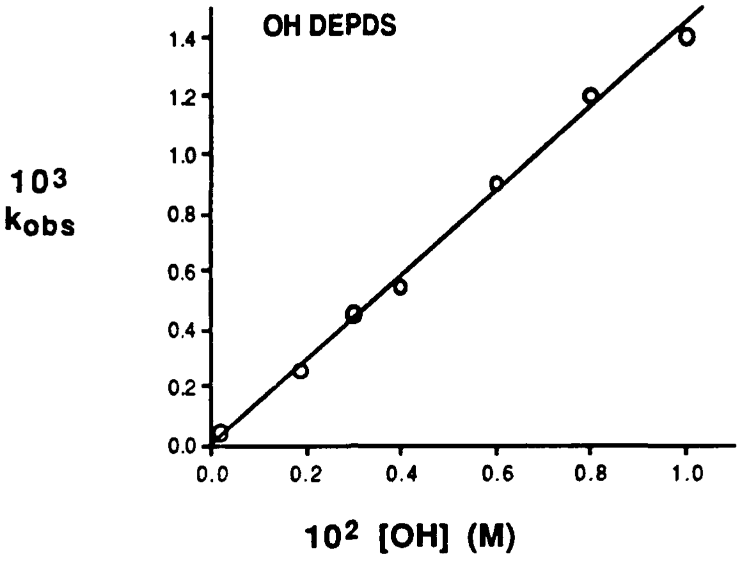

Figure 5. A Plot of k_{obsd} vs $[OH^-]$ in the Absence of IBA. The slope equals k_{OH}, since $k_{hyd} \sim 0$.

Table 2. Values of the Rate Constants in Aqueous Borate Buffer Solutions, at 25 °C

pH	$k_{hyd} + k_{OH}\,[OH^-]$ / s^{-1}	k_{IBA} / $s^{-1}\,M^{-1}$
8.5	$(5.38 \pm 0.22) \times 10^{-5}$	0.519 ± 0.021
9.2	$(6.28 \pm 0.46) \times 10^{-5}$	0.675 ± 0.038
10.4	$(2.03 \pm 0.06) \times 10^{-4}$	0.798 ± 0.045

Table 3. Effect of OH^- and k_{obsd} in Aqueous Solution in the Absence of IBA, at 25 °C

10^2 [OH^-]	10^3 k_{obsd} / s^{-1}
1.00	4.16 ± 0.03
0.75	3.17 ± 0.01
0.50	1.98 ± 0.01
0.25	0.97 ± 0.03

In order to demonstrate unambiguously that IBA is indeed a turnover (rather than stoichiometric) catalyst in the CTAB microemulsion medium two additional experiments were performed. In the first of these, the PNDP concentration was increased such that the ratio of PNDP to IBA was 115 to 1 at a pH of 9.5. In order to study this reaction spectrophotometrically, data were taken at 470 nm where the extinction coefficient of 4-nitrophenoxide is only 763 $M^{-1}cm^{-1}$. Normal first-order kinetics were observed with a calculated rate constant consistent with equation 6. In addition, if IBA were consumed in the reaction it would be quickly depleted and limited rate enhancement would be observed. (It should be pointed out that we did not observe "burst kinetics" in this or any other experiment.[5a])

The second experiment involved repeated addition of PNDP to the same reaction mixture. In the first step, PNDP was added to the microemulsion containing IBA and the kinetics carefully followed. After ten half-lives, a second addition of PNDP was added to the same solution and kinetics were again followed. The first-order rate constants obtained from both segments of the reaction were identical within experimental error, indicating that the first reaction had no effect on the IBA concentration.

Table 4. Effect of CTAC on k_{OH} and k_{IBA} in Aqueous Micellar Solution, at 25 °C

CTAC M^{-1}	$k_{hyd} + k_{OH}[OH^-] \times 10^{-3}$ s^{-1}	10^{-3} k_{IBA} s^{-1} M^{-1}
0.82	2.20 ± 0.02	1.55 ± 0.05
1.22	2.04 ± 0.15	1.45 ± 0.06
2.03	2.00 ± 0.01	1.13 ± 0.02
2.82	1.96 ± 0.01	1.10 ± 0.02

4 CONCLUSION

The introduction of IBA into the CTAB microemulsion increases the reaction rate by a factor of 10 above background hydrolysis. In addition, the catalyzed rate in CTAB microemulsion is approximately twice the value in water. There is no doubt that IBA functions as a true catalyst; in our experiments we obtained an average turnover number of 25 with no indication of loss of catalytic behavior in CTAB microemulsions. We should indicate that none of our observations are inconsistent with the mechanisms which have previously been proposed for either the catalyzed or the uncatalyzed reaction.[5,8,9]

Examination of the results of our studies and of the studies reported in the literature proves that the CTAC and CTAB micellar solutions are truly remarkable media. Second-order rate constants for the hydrolysis catalysts IBA and its derivatives are at least 1000 times greater in these micellar solutions than in any of the other media studied.[4,5] Further studies of these hydrolysis reactions should be aimed at determining the structural feature(s) of micellar solutions which make them catalytic media. Perhaps then it will be possible to combine the good solubilization properties of microemulsions with the catalytic properties of micellar solutions to produce an ideal medium.

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