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# The Mechanism of Dephosphorylation of Bis(2,4-dinitrophenyl) Phosphate in Mixed Micelles of Cationic Surfactants and Lauryl Hydroxamic Acid

Marcelo Silva,[†] Renata S. Mello,[†] M. Akhyar Farrukh,[†] Janio Venturini,[†] Clifford A. Bunton,[‡] Humberto M. S. Milagre,[§,∥] Marcos N. Eberlin,[§] Haidi D. Fiedler,[†] and Faruk Nome*,[†]

[†]*Departamento de Química, Universidade Federal de Santa Catarina, 88040-900 Florianópolis, SC, Brazil,* [‡]*Department of Chemistry and Biochemistry, University of California, Santa Barbara, California 93106, and* [§]*ThoMSon Mass Spectrometry Laboratory, Institute of Chemistry, University of Campinas, UNICAMP, 13083-970 Campinas, SP, Brazil.* [∥]*Current address: Department of Biochemistry and Microbiology, UNESP - São Paulo State University, 13506-900 Rio Claro, SP, Brazil.*

*faruk@qmc.ufsc.br*

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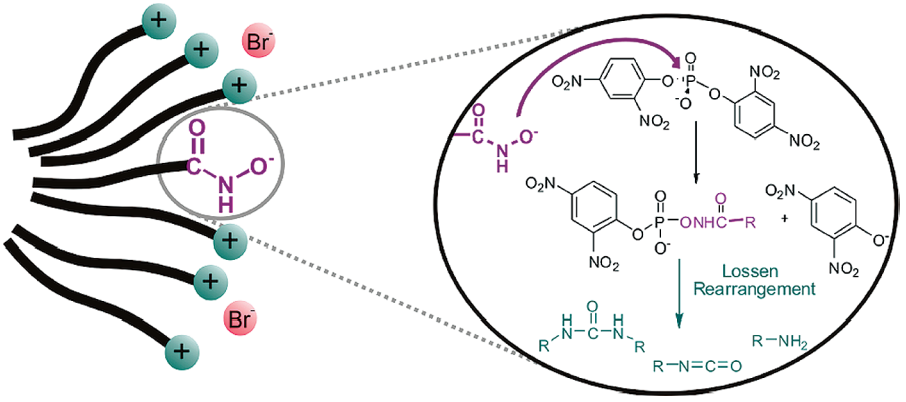


Mixed micelles of cetyltrimethylammonium bromide (CTABr) or dodecyltrimethylammonium bromide (DTABr) and the α-nucleophile, lauryl hydroxamic acid (**LHA**) accelerate dephosphorylation of bis(2,4-dinitrophenyl)phosphate (**BDNPP**) over the pH range 4−10. With a 0.1 mole fraction of **LHA** in DTABr or CTABr, dephosphorylation of **BDNPP** is approximately $10^4$-fold faster than its spontaneous hydrolysis, and monoanionic $\mathbf{LHA}^-$ is the reactive species. The results are consistent with a mechanism involving concurrent nucleophilic attack by hydroxamate ion (i) on the aromatic carbon, giving an intermediate that decomposes to undecylamine and 2,4-dinitrophenol, and (ii) at phosphorus, giving an unstable intermediate that undergoes a Lossen rearrangement yielding a series of derivatives including *N*,*N*-dialkylurea, undecylamine, undecyl isocyanate, and carbamyl hydroxamate.

## Introduction

Reactions such as hydrolysis and nucleophilic substitution on phosphoesters are involved in important biological processes,[1] and extensive studies have been developed in attempts to understand such crucial reactions.[1−4] Monoanionic phosphodiesters are generally less reactive than the corresponding phosphomonoesters; for example, the relatively slow spontaneous hydrolysis of monoanionic bis-2,4-dinitrophenyl phosphate, **BDNPP** (**1**), initially forms the 2,4-dinitrophenyl phosphate monoester dianion (**DNPP**) with P−O bond fission, and the initial reaction is pH independent at 4−7,[5,6] but the spontaneous hydrolysis of **DNPP** is over 1000 times faster than that of monoanionic **BDNPP**.[5,6]

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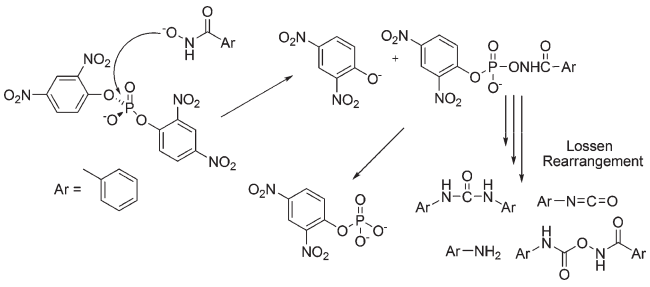
**SCHEME 1. Self-Destruction of Benzohydroxamic Acid via Lossen Rearrangement**




However, alkaline hydrolysis of **BDNPP** is much faster than that of **DNPP**, which complicates development of efficient reagents that fully degrade phosphodiesters because initially formed phosphate monoester dianions are unreactive to anions due to the considerable electrostatic barrier to nucleophilic attack.

α-Effect nucleophiles such as hydroxylamine,[7] hydrazine,[8] and hydroxamic acid[4,9,10] are especially effective dephosphorylating agents. In an earlier investigation, products and intermediates of dephosphorylation of **BDNPP** anion by benzohydroxamate anion (**BHO**$^-$) were identified, showing that the reaction follows parallel paths with hydroxamate attack on phosphorus and on the aromatic carbon.[4] The attack on carbon gives an intermediate, which was detected, but slowly decomposes to aniline and 2,4-dinitrophenol, whereas attack on phosphorus gives an unstable intermediate that undergoes a Lossen rearrangement to urea, amine, isocyanate, and carbamyl hydroxamate.[11−16] Thus, **BHO**$^-$ behaves as a self-destructive intramolecular scissor which reacts and loses nucleophilic ability (Scheme 1).

Reactivities of hydroxamate ions are increased by incorporation in functional polymers or by comicellization with inert surfactants in water.[17−19] As expected on the basis of pseudophase treatments of micellar rate effects,[20,21] high local concentrations of hydroxamate ions in the interfacial micellar region are largely responsible for these rate increases.[18] Considering that (i) cationic micelles of cetyltrimethylammonium bromide, **CTABr**, accelerate the spontaneous hydrolysis of **BDNPP** up to 30-fold and high pH accelerates the reaction by concentrating nucleophiles, e.g., $OH^-$,[21,22] and (ii) cationic micelles accelerate the reaction of hydroxamates and phosphate triesters,[18,19,21] we examined micellar effects on reactions of the less reactive phosphodiesters with simple hydroxamates. We selected lauryl hydroxamate because its long alkyl chain should facilitate mixed micelle formation, effectively increasing nucleophilic concentration in the micellar pseudophase, and accelerating dephosphorylation of **BDNPP** by the α-nucleophile. We monitored via several spectroscopic and mass spectrometric methods the dephosphorylation products of **BDNPP** in comicelles of the amphiphilic α-nucleophile, lauryl hydroxamic acid, as a functional surfactant, and dodecyl- or cetyltrimethylammonium bromide.

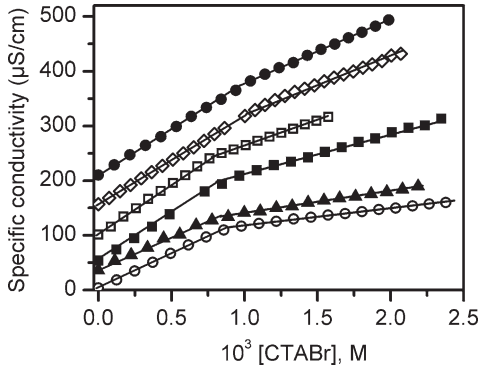



**FIGURE 1.** Specific conductance as a function of [CTABr] in the absence of **LHA** (○) and in the presence of molar fractions of **LHA** of 0.048 (▲), 0.091 (■), 0.11 (□), 0.14 (◊), and 0.20 (●), at 25.0 °C, in the absence of buffer, pH 10.0.

## Results and Discussion

**Micelle Formation and Fractional Ionization.** Critical micelle concentrations, CMC, and fractional micellar ionizations, α, for mixed micelles of **LHA** and CTABr were estimated from plots of conductance against [surfactant], Figure 1. Breaks in these plots give CMC values that change little with increasing $\chi_{LHA}$ but approximate values of α, from the ratio of slopes,[23−26] increase with increasing $\chi_{LHA}$, indicating that addition of dodecyl hydroxamate ion effectively neutralizes the cationic micelles, decreasing the micellar surface potential and affinity for counteranions. This effect is very sensitive to $\chi_{LHA}$ (Table 1).

The CMC of DTABr in water is $1.5 \times 10^{-2}$ M, but for a mixture of DTABr and **LHA** $\chi_{LHA} = 0.1$, the CMC decreases to $3 \times 10^{-3}$ M, and the fractional micellar ionizations, α, sharply increase from 0.27 to 0.73; i.e., **LHA** significantly increases the fractional micellar charge of both CTABr and

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**TABLE 1. Critical Micelle Concentrations and α at Different $\chi_{LHA}$ in CTABr/LHA Mixtures**[a]

| $\chi_{LHA}$ | α | $10^4$ CMC, M |
|---|---|---|
| 0.0 | 0.23 | 8.8 |
| 0.048 | 0.41 | 7.9 |
| 0.091 | 0.43 | 7.7 |
| 0.11 | 0.50 | 6.8 |
| 0.14 | 0.60 | 9.9 |
| 0.20 | 0.71 | 8.3 |

[a]At pH = 10.0, 25 °C, 0.01 M.

DTABr depending on both $\chi_{LHA}$ and chain length of the cationic surfactant. Decreases in the CMC are larger with the shorter chain DTABr, and the fractional micellar charge of CTABr tends toward that of pure surfactant as $\chi_{LHA}$ approaches zero.

**Kinetic Studies in Cationic Micelles.** The relatively slow spontaneous hydrolysis of **BDNPP** at pH 8.0 ($k = 2.5 \times 10^{-7}$ $s^{-1}$) is accelerated ca. 30-fold by cationic micelles of CTABr.[27] The results for hydrolysis of **BDNPP** at pH 10.0 (Figure 2) are qualitatively similar to those reported by Buist et al.[27] and are as expected for reaction of hydroxide ion and a strongly micellar bound organic substrate.[20] As is typical of bimolecular reactions, $k_{obs}$ increases to maxima and decreases as reagents are diluted due to increasing micellar volume.[20]

**Kinetic Studies in Mixed Micelles.** The dephosphorylation of **BDNPP** at pH 10.0, (self-buffered, see the Experimental Section) in mixed micelles of CTABr or DTABr and lauryl hydroxamic acid (**LHA**) is much faster than in cationic surfactant. The absorbance increase at 400 nm is consistent with a yield of 200 mol % of **DNP**,[28] although another product absorbing at ca. 360 nm formed more rapidly in a complex two-step reaction (see the Supporting Information).[4]

**Product Characterization.** As shown earlier,[4] dephosphorylation of **BDNPP** anion by benzohydroxamate anion (**BHO**$^-$) follows two reaction paths: (i) reaction at phosphorus, yielding an unstable intermediate that undergoes a Lossen rearrangement to phenyl isocyanate, aniline, diphenylurea, and *O*-phenylcarbamyl benzohydroxamate, and (ii) attack on the aromatic carbon giving an intermediate, which was detected, but slowly decomposes to aniline and 2,4-dinitrophenol.[4] To investigate whether the reaction of **LHA** and **BDNPP** in a cationic micelle follows the same paths, ESI-MS in the negative mode was used to monitor the course of reaction by collecting snapshots of the anionic composition of the reaction solution.[29] Reactants, intermediates, and anionic products were transferred directly from the reaction solution to the gas phase and then detected by ESI(−)-MS. Figure 3 shows two characteristic ESI-MS collected at the same reaction time but with different pH.

The ESI(−)-MS of Figure 3 detected several anions: **BDNPP** of *m*/*z* 429, **LHA**$^-$ of *m*/*z* 214, and anions from the cleavage of **BDNPP** such as the phenoxide **DNP**, **5**, of *m*/*z* 183, $\mathbf{H_2PO_4^-}$ of *m*/*z* 97, $\mathbf{PO_3^-}$ of *m*/*z* 79, and $\mathbf{H_2PO_3^-}$ of *m*/*z* 81. The most mechanistically relevant anion is, however, that of *m*/*z* 460, which confirms the hydroxamate attack on phosphorus (path A in Scheme 2). The structural assignment of this crucial intermediate was confirmed by ESI(−)-MS/MS, through which it was shown to dissociate to form phenoxide **DNP**, **5**, of *m*/*z* 183 (see the Supporting Information). The attack via path A in Scheme 2 forms the expected cleavage products of **BDNPP**,[28] the phenoxide **5**, and intermediate **4** of *m*/*z* 460, which subsequently generates **6** and other products as discussed later. The fragment anion of *m*/*z* 167 may come from oxygen loss in **5**. The kinetics of the reaction of **LHA**$^-$ and **BDNPP** are typical of two concurrent reactions where, besides attack on phosphorus, a compound absorbing in the UV is formed, which is consistent with attack on the aromatic carbon (path B in Scheme 2). The attack on carbon gives intermediate **7**, which subsequently decomposes via a Lossen rearrangement to **DNP**, undecylamine, **9**, and other products discussed below.

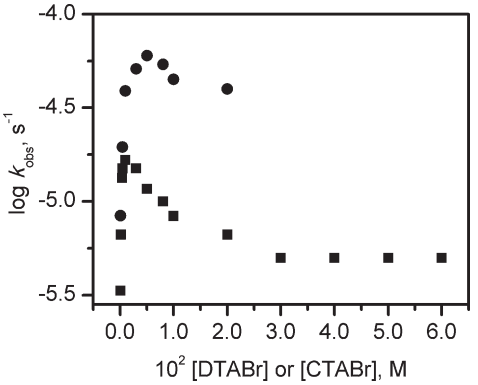


**FIGURE 2.** Observed rate constant for the hydrolysis of **BDNPP** as a function of [DTABr] (■) or [CTABr] (•), pH = 10.0, at 25 °C in borate buffer 0.01 M.

Reaction products from the reaction of **LHA** and **BDNPP** were identified by absorption and NMR spectra. Comparison was made with those of authentic material, with **LHA** in significant excess over the sodium salt of **BDNPP**, which was used to avoid $^1$H signals of the pyridinium ion which would complicate the $^1$H NMR spectra.[28] Unfortunately, aliphatic $^1$H NMR signals were not useful because they are obscured by signals of the excess **LHA**, and H−D exchange precludes examination of the NH signals. After complete reaction, only 2,4-dinitrophenoxide ion was identified by absorption and NMR spectroscopy (δ 6.74 (d, 1H, $J$ = 9.6 Hz, Ar), 8.11 (dd, 1H, $J_{ab}$ = 9.6 Hz and $J_{bx}$ = 3.0 Hz, Ar), 8.90 (d, 1H, $J$ = 3.0 Hz, Ar)), and the signals were identical with those of 2,4-dinitrophenol at the appropriate pH. The signal of inorganic phosphate (δ 1.86 ppm) failed to appear simultaneously with **DNP**, indicating formation of phosphorylated lauryl hydroxamic acid derivatives, which slowly decompose, similar to the reaction products of **BDNPP** and hydroxylamine.

Electron ionization mass spectrometry (EI-MS) with the direct injection mode was also applied to monitor the formation of less polar, nonionic products, and it was successful in identifying other important products such as those from the Lossen rearrangement (see the Supporting Information). These products were extracted with ether from the reaction products of **BDNPP** with aqueous **LHA**$^-$ (1:1) at pH 9, 25 °C after 10 min. EI-MS data were consistent with formation of 1-isocyanatoundecane (**8**, *m*/*z* 197), undecylamine (**9**, *m*/*z* 171), and 1,3-diundecylurea (**10**, *m*/*z* 368). The

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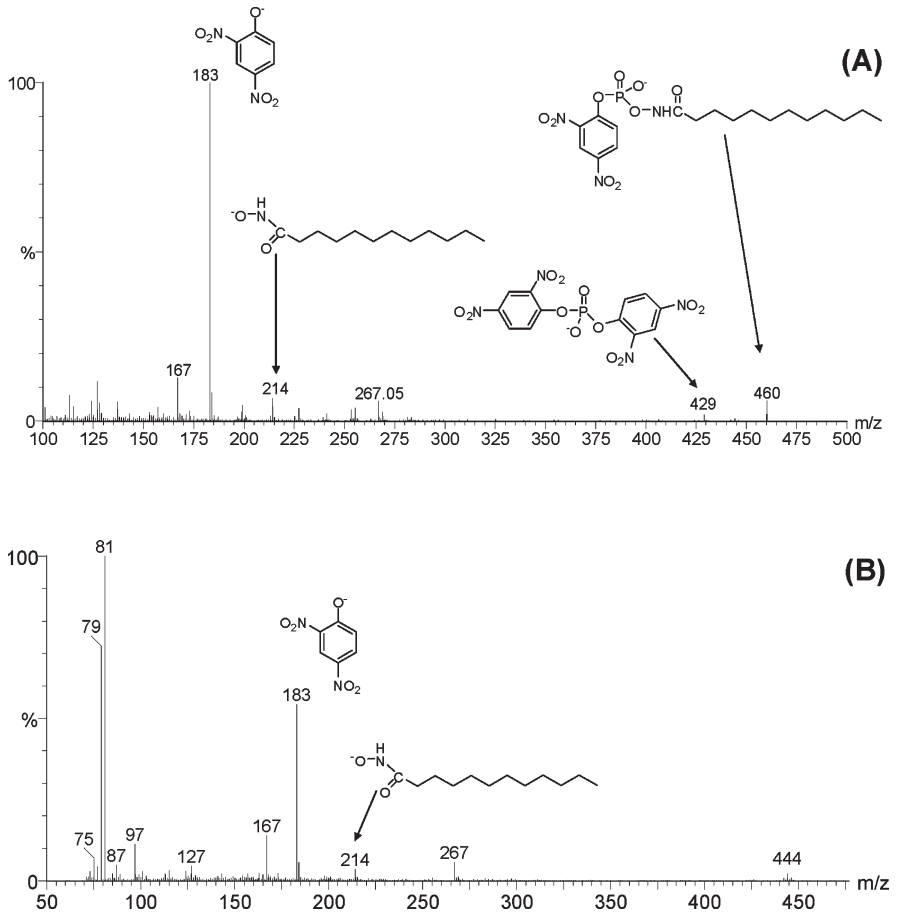
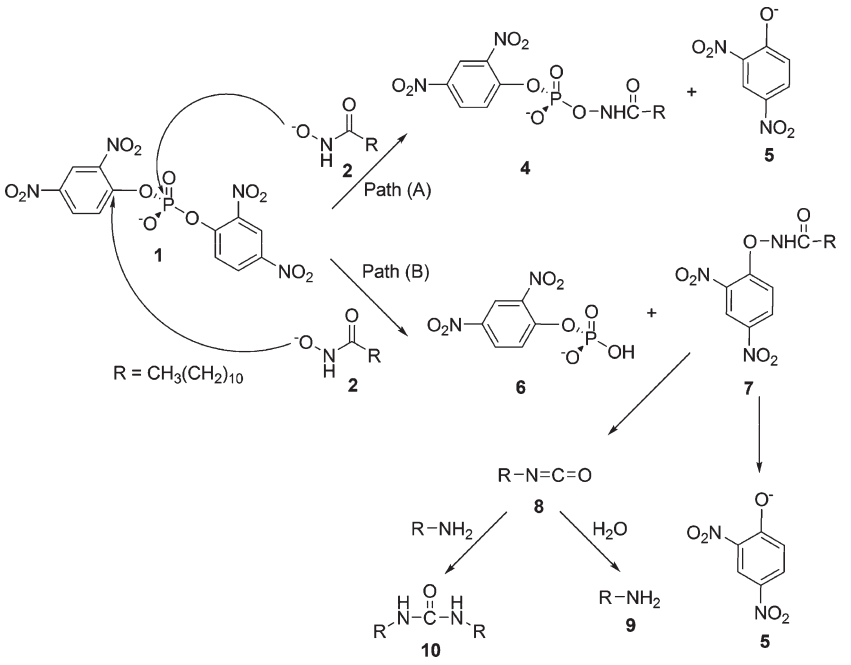


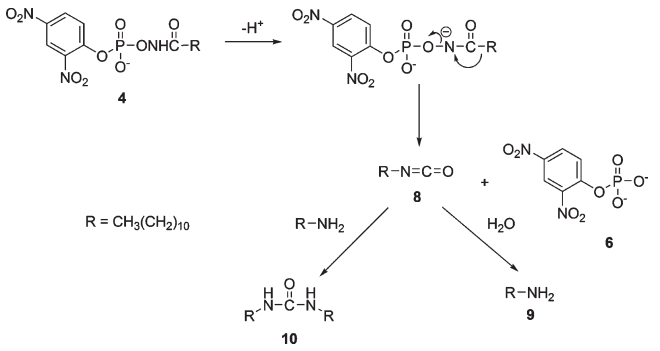

**FIGURE 3.** ESI(−)-MS for the reaction solution of **LHA/CTABr** and **BDNPP** at 25 °C, (A) pH 8 and (B) pH 9.

**SCHEME 2. Nucleophilic Attack of $LHA^-$ by Two Reaction Paths: (Path A) at Phosphorus and (Path B) on the Aromatic Carbon**




results are consistent with Scheme 3 and indicate that the *O*-phosphorylated intermediate **4**, in basic media, decomposes to **DNPP** and undergoes a Lossen rearrangement to give 1-isocyanatoundecane (**8**) in a rearrangement similar to that of intermediate **7**. Subsequently, the isocyanate **8** decomposes to undecylamine **9**, which reacts with isocyanate **8**,

**SCHEME 3. Self-Destruction of the Intermediate from the Reaction of LHA$^-$ and BDNPP via a Lossen Rearrangement**




giving urea **10**. Moreover, the EI-MS data shows ions of $m/z$ 325, 311, 297, 283, 269, and 255 that may be related to consecutive losses of 28 Da from ionized 1,3-diundecylurea (**10**) a normal EI-MS pattern for long-chain dialkylureas.[30] Scheme 3 shows this rearrangement and as for the benzohydroxamate anion, lauryl hydroxamate anion also behaves as a self-destructive nucleophile because it loses its nucleophilicity by reacting effectively with **BDNPP**. The concentration of 2,4-dinitrophenoxide ion (DNP) formed in the first step of the reaction was used to estimate that the reaction proceeds with ca. 83% of attack on the phosphorus atom.

**Detailed Kinetic Studies of the Reaction of BDNPP and LHA$^-$.** At constant $\chi_{LHA}$ and self-buffered, pH 10.0, rate constants are much higher than in the absence of **LHA** (Figure 4). With both CTABr and DTABr and $\chi_{LHA}$ = 0.1, $k_{obs}$ is independent of [surfactant], indicating that **LHA** and the substrate are entirely in the micellar phase with the constant mole fraction of **LHA**. The rate constants for the **LHA** reaction are similar in CTABr and DTABr, and the average value is 0.021 $s^{-1}$, which is several orders of magnitude greater than that in water and reaction is 350- and 1200-fold faster than the spontaneous reaction in DTABr and CTABr, respectively.

Figure 5 illustrates the pH−rate profile for the dephosphorylation of **BDNPP** by **LHA**, in the presence of DTABr and CTABr, where $k_{obs}$ increases with pH and reaches a plateau in the pH region where **LHA** is fully deprotonated. The spontaneous hydrolysis of **BDNPP** is included for comparison and at pH 10, $k_{obs}$ is about $9 \times 10^{-7}$ $s^{-1}$ for this reaction. Note the striking acceleration of the reaction, ca. $2 \times 10^4$ fold, with micellar incorporation of **LHA$^-$** at pH 10. This acceleration is due to (i) hydrophobic and charge effects which concentrate reagents in the mixed micelle and (ii) the nucleophilicity of monoanionic lauryl hydroxamate, which is an effective α-effect nucleophile with unshared electron pairs on oxygen and nitrogen.

These micellar effects are treated in terms of equilibrium distributions of **BDNPP** and hydroxamate between the aqueous and micellar pseudophases (Scheme 4), where subscripts m and w indicate micellar and aqueous pseudophases, respectively.[20]

The rate constants in the presence of **LHA** are significantly greater than that in its absence, and under our experimental conditions, both **BDNPP** and **LHA** are fully incorporated in the micellar phase (see Figure 4). Hence, we can neglect reactions in water and in the absence of **LHA** and treat $k_{obs}$ as a function of pH by using eq 1, which relates rate constants and hydroxamate formation, in terms of pH and the apparent p$K_a$ of **LHA**. The formalism of eq 1 gives an apparent dissociation constant of 9.2 for **LHA** in CTABr/DTABr, at constant $\chi_{LHA}$ = 0.1, and a rate constant in the plateau region of 0.025 $s^{-1}$.

$$k_{obs} = k_m \chi_{LHA} \frac{K_a}{(K_a + [H^+])} \quad (1)$$

As expected, $k_{obs}$ follows $\chi_{LHA}$ and Figure 6 shows that increase of [**LHA**] at constant [quaternary ammonium surfactant] significantly increases $k_{obs}$ for the dephosphorylation of **BDNPP**.

The simple form of eq 1 results from total micellar incorporation of the substrate, which is confirmed by the generally constant $\chi_{LHA}$. The data in Figure 5 give the second-order rate constant in mole fraction units as $k_m$ = 0.25 $s^{-1}$. This second-order rate constant can be compared with that in water, which is written as $M^{-1}$ $s^{-1}$, by assuming a

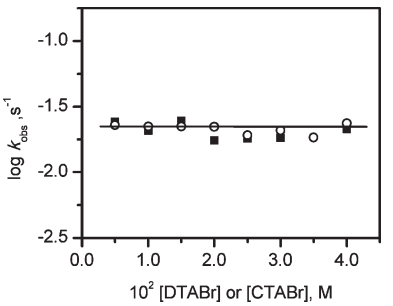



**FIGURE 4.** Observed rate constant for the dephosphorylation of **BDNPP** ($1.33 \times 10^{-5}$ M) in the presence of **LHA**, as a function of [DTABr] (■) or [CTABr] (○), pH = 10.0, without added buffer, 25 °C, and constant ([LHA]/[surfactant]) = 0.1.

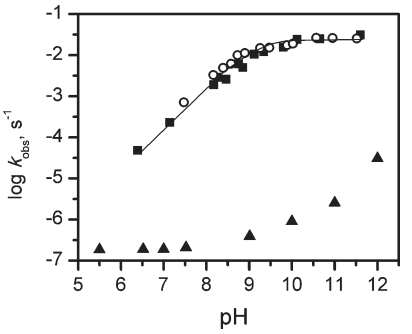



**FIGURE 5.** Dependence of log $k_{obs}$ on pH for the reaction of **BDNPP** in water[5] (▲) and in the presence of $1.0 \times 10^{-2}$ M (■) DTABr or (○) CTABr and $1.0 \times 10^{-3}$ M **LHA**, in the absence of buffer, at 25 °C.

**SCHEME 4**

$$\mathrm{BDNPP_w} + \mathrm{Nu_w} \xrightarrow{k_w} \text{Products}$$

$$\mathrm{BDNPP_m} + \mathrm{Nu_m} \xrightarrow{k_m} \text{Products}$$

(30) SDBS Web: http://riodb01.ibase.aist.go.jp/sdbs/ (National Institute of Advanced Industrial Science and Technology, Acessed: May 19, 2009).

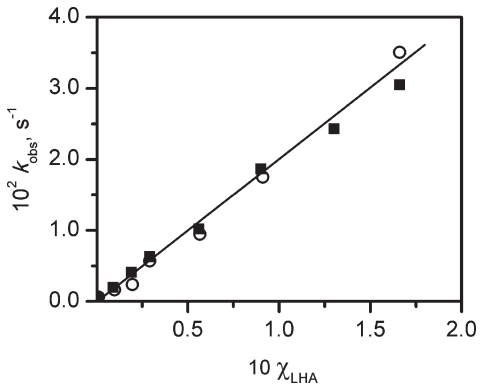



**FIGURE 6.** Effect of $\chi_{LHA}$ on $k_{obs}$ ($s^{-1}$) for reaction of **BDNPP**, pH = 10, without buffer with (■) [DTABr] or (○) [CTABr] = $1 \times 10^{-2}$ M, in aqueous solution at 25 °C.

value for the molar volume of the micellar reaction, which is uncertain, but is probably ca. 0.2 $M^{-1}$, giving a rate constant ($k_{2obs}$) of ca. 0.05 $M^{-1}\ s^{-1}$. Based on the percentage of attack on the aromatic ring and on the phosphorus atom (see above), the constant $k_{2obs}$ was separated into two constants, $k^{C}_{LHA-} = 8.5 \times 10^{-3}\ M^{-1}s^{-1}$and $k^{P}_{LHA-} = 4.15 \times 10^{-2}\ M^{-1}\ s^{-1}$, that correspond to the attack on the aromatic ring and phosphorus, respectively.

The second-order rate constants for reaction of a similar alkyl hydroxamate ion in water are not known, but for reaction with benzohydroxamate ion[4] the values of the second-order rate constants for attack on the aromatic ring and phosphorus are $k^{C}_{BHO-} = 1.98 \times 10^{-1}\ M^{-1}s^{-1}$and $k^{P}_{BHO-} = 6.21 \times 10^{-2}\ M^{-1}\ s^{-1}$, respectively. The experimental results are indicative that, as is typical of reactions of anionic nucleophiles, micellar accelerations are due, to a first approximation, to concentration of the two reactants in the small volume of the micellar reaction region.

## Conclusions

Mixed micelles of **LHA** and either CTABr or DTABr effectively accelerate dephosphorylation of **BDNPP** by concentrating the α-nucleophile in the micellar pseudophase. The rate constants increase as a function of pH. Formation of the reactive lauryl hydroxamate and the observed rate constant is linearly dependent on the nucleophilic mole fraction in the micellar pseudophase, and when it is 0.1, dephosphorylation of **BDNPP** is approximately $10^4$-fold faster than spontaneous hydrolysis. The reaction was monitored by ESI-MS and EI-MS, and results obtained for both polar and less polar species are consistent with a mechanism involving simultaneous nucleophilic attack of the hydroxamate (i) at phosphorus, giving an unstable intermediate that undergoes a Lossen rearrangement forming urea, amine, isocyanate, and carbamyl hydroxamate, which were detected, and (ii) on the aromatic carbon, giving an intermediate which was detected but decomposed to undecylamine and 2,4-dinitrophenol.

The nucleophilicity of **LHA** and **BHO** (using **BDNPP** as a model substrate) is similar to that of a series of oximes and hydroxylamines which show reasonable activity as DNA-cleavage agents, and these compounds can be coupled to peptides or other molecules to promote targeting to specific DNA sequences.[31] Thus, similarly to results previously reported, **LHA** behaves as a self-destructive intramolecular scissor and loses its nucleophilicity after reacting with the phosphate diester. This characteristic of the nucleophile could be relevant in the design of special scissors for gene therapy.

## Experimental Section

**Materials. BDNPP** as the pyridinium salt was prepared as described.[5] The pyridinium ion was exchanged for $Na^+$ on the cation-exchange resin in the $Na^+$ form. The potassium salt of lauryl hydroxamic acid was prepared from ethyl laurate following a described procedure[32] and ethyl laurate was synthesized from lauric acid by a known procedure.[33] Lauryl hydroxamic acid (**LHA**) was synthesized by modifying a described method,[32] yielding 74% of a white solid, with mp in agreement with the literature (92 °C).[34,35] All of the modifications in the syntheses are described in the Supporting Information. All other inorganic reagents and 2,4-dinitrophenol were of the highest purity available and were used as purchased.

**Kinetics.** Reactions followed spectrophotometrically were started by adding 10 μL of a stock solution of the substrate ($4 \times 10^{-3}$ M) in MeCN to 3 mL of the reaction mixture, containing a large excess of the nucleophilic hydroxamate, assuring strictly first-order kinetics for the initial nucleophilic attack upon the substrate. Solutions were self-buffered by **LHA/LHA$^-$** and were prepared by addition of aqueous standard 0.1 M NaOH to aqueous **LHA**. Reaction pH in aqueous solutions, without **LHA**, was controlled by borate buffer (0.005 M) from pH 8 to 10 and by NaOH at higher pH.[4]

Reactions were followed, at 25.0 ± 0.1 °C, by appearance of **DNP** at 400 nm on a diode-array spectrophotometer with a water-jacketed thermostated cell holder. The pH of each reaction mixture was measured at the end of each run and verified that the pH remained constant during the reaction. Observed first-order rate constants ($k_{obs}$) correspond to the initial reaction of **BDNPP** and were calculated from linear plots of $\ln(A_\infty - A_t)$ against time for at least 90% of the reaction by using and interactive least-squares program; correlation coefficients were > 0.999 for all kinetic runs.

**Mass Spectrometry.** In order to identify intermediates and reaction products of **BDNPP** with **LHA$^-$**, direct infusion ESI-(−)-MS analyses was performed with an instrument with 5.000 mass resolution and 50 ppm mass accuracy in the TOF mass analyzer. The typical operating conditions were as follows: 3 kV capillary voltage, 8 V cone voltage, and desolvation gas temperature of 100 °C. Tandem ESI-MS/MS was collected after 4−32 eV collision induced dissociation (CID) of mass-selected ions with argon. Mass-selection was performed by Q1 using a unitary $m/z$ window, and collisions were performed in the rf-only quadrupole collision cell, followed by mass analysis of product ions by the high-resolution orthogonalreflectron TOF analyzer operated in the negative-ion mode.[3,28] For typical electrospray ionization (ESI-MS) conditions, 1 mL of $1 \times 10^{-6}$ M **BDNPP**, in aqueous medium at pH 8 and 9, was mixed with 100 μL $min^{-1}$ of 0.1 M aqueous **LHA$^-$/CTABr**. A microsyringe

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pump delivered the reagent solution into the ESI source at a flow rate of 10 $\mu$L $min^{-1}$. Main ESI(−)-MS conditions were as follows: curtain gas nitrogen flow of 20 mL $min^{-1}$; ion spray voltage of −4500 eV; declustering potential of −21 eV; entrance potential of −10 eV; collision cell exit potential of −12 eV. Some of the main anionic species detected by ESI(−)-MS were subjected to ESI-MS/MS by using collision-induced dissociation (CID) with nitrogen and collision energies ranging from 5 to 45 eV.

EI-MS analyses were also performed to detect less polar, nonionic products in the reaction medium. Reaction of **BDNPP** with **LHA**$^-$ (1:1) was followed in aqueous solution at pH 9 and 25 °C for 10 min, and the products were extracted with diethyl ether and after evaporation. The EI-MS data from the mixture was obtained by using the direct injection mode, with EI at 70 eV. The samples were placed in a sample vial fixed onto the probe. Probe temperatures were programmed as follows: 10 °C.$min^{-1}$ up to 85 °C and held for 3 min, increasing from 85 to 300 at 20 °C.$min^{-1}$, and held for 3 min.

**NMR Spectroscopy.** All the $^1$H and $^{31}$P NMR spectra were monitored on a spectrometer (400 MHz for $^1$H) in $D_2O$ at 25 °C and, because relaxation is slow at some nitroarene positions, generally with a delay time of 20 s.[7] The $^1$H chemical shifts are referred to internal sodium 3-(trimethylsilyl)propionate (TSP), and those of $^{31}$P are referred to external 85% phosphoric acid. The value of pD was obtained by adding 0.4 to the observed pH of solutions in $D_2O$ at 25 °C.[36]

**Potentiometric Titration.** Potentiometric titrations were carried out with a pH meter and an automatic buret, in a 150.0 mL thermostated cell, under $N_2$ at 25 °C. A solution, of 20.0 mL of 2.0 mM of **LHA** and 20 mM of CTABr was acidified with 1.0 mL of 0.1 M HCl and titrated with small increments of $CO_2$ free 0.1008 M KOH with 0.1 ionic strength (KCl). The FITEQL v.4.0[37] was used to calculate the constants.

**Conductance Measurements.** Conductance measurements were carried out at 25.0 ± 0.1 °C in a water-jacketed flow dilution cell with a bridge-type conductimeter. Values of the CMC were determined in the usual manner from plots of specific conductance against surfactant concentration. Solutions were self-buffered by **LHA/LHA**$^-$. The pH was measured in a thermostated stirred vessel with a pH meter. The glass electrode was calibrated against standard buffers, at 25.0 ± 0.1 °C.

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**Supporting Information Available:** Preparation of ethyl laurate, potassium salt of lauryl hydroxamic and lauryl hydroxamic acid (LHA), UV absorption spectra for the reaction of **LHA**$^-$ with **BDNPP**, ESI-MS/MS of the key intermediate on $m/z$ 460, EI-MS data for product identification, and kinetic data for reaction of **BDNPP** with **LHA**$^-$. This material is available free of charge via the Internet at http://pubs.acs.org.

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