Co-operation of Two Amino Residues for the Facile Cleavage of Bis(nitrophenyl) Phosphates

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N,N,N',N'-Tetramethylalkanediyldiamines and their monocations are effective catalysts for the cleavage of bis(2,4-dinitrophenyl) and bis(4-nitrophenyl) phosphates.

The preparation of artificial nucleases has attracted much interest.[1] Most of the systems hitherto reported involve metal complexes as the catalytic sites for the cleavage, which are attached to the moieties showing molecular recognition for specific positions of nucleic acids. Attempts to find small organic molecules which will serve as the catalytic sites have not been sufficiently successful yet. Poly(amino acid)s[2] were found to be applicable only to the cleavage of ribonucleic acids.

We now report that *N,N,N',N'*-tetramethylalkanediyldiamines ($Me_2N[CH_2]_nNMe_2$: n = 1, N–1–N; n = 2, N–2–N; n = 3, N–3–N) and their monocations exhibit significant catalysis for the cleavage of bis(2,4-dinitrophenyl) phosphate (**1**) and bis(4-nitrophenyl) phosphate (**2**). The catalytic activities are enormously enhanced by the co-operation of the two amino residues (either cationic or neutral). Furthermore, the attachment of an ethylenediamino residue to β-cyclodextrin as a substrate-binding site provides an even more effective catalyst having activity comparable to that of active metal complex catalysts.

The cleavage at 50 °C was followed at 400 nm. The second-order catalytic rate constant k was determined from the slope of the linear plot of the pseudo-first-order rate constant *vs.* the concentration of the corresponding species (evaluated using the titrimetrically determined pK_{a1} and pK_{a2} values). The β-cyclodextrin bearing an *N,N*-dimethylethylenediamino residue at the primary hydroxy side (CyD-N-2-N) was prepared by the reaction of 6-*O*-monotosyl-β-cyclodextrin with *N,N*-dimethylethylenediamine.

The cleavage of (**1**) is efficiently catalysed by the monocations of N–1–N, N–2–N, and N–3–N, as shown in Table 1 (450-fold acceleration by 1.0 M N–1–N at pH 8.0). In contrast, trimethylamine (NMe_3) in the neutral form shows only a marginal catalysis. The closely separated $-NMe_2H^+$ residues in the monocations function as general acid catalysts, promoting the nucleophilic attack[3] of the $-NMe_2$ residue, as depicted in Figure 1(a), by more than 870, 74, and 190 fold for N–1–N, N–2–N, and N–3–N. The general acid mechanism is supported by the significant D_2O solvent isotope effect (k_H/k_D 1.6 for N–1–N). The possibility that the $-NMe_2H^+$ residues simply increase the local concentration of (**1**) around the catalysts by electrostatic attraction is ruled out, since the catalysis by *N,N,N,N',N'*-pentamethylethylenediamine is much smaller ($k = 1.2 \times 10^{-4}$ mol^{-1} dm^3 min^{-1}).

Figure 1. Proposed mechanisms for the catalysis by (a) the monocations and (b) the neutral diamines. R represents the nitrophenyl residue, and the methyl groups on the nitrogen atoms are omitted for clarity.

Furthermore, the neutral N–1–N, N–2–N, and N–3–N species also surpass NMe_3 in catalytic activity by more than 1800—2000 fold (Table 1). Large D_2O solvent isotope effects were not observed. Here, the lone-pair electrons on the second nitrogen atom electrostatically stabilize the positive charge, formed on the first nitrogen atom by nucleophilic attack [Figure 1(b)].

The acceleration of the cleavage of (**2**) by N–1–N is still more striking: 3900 and 2400 fold at pH 7.5 and 11.0, respectively. NMe_3 shows virtually no catalytic activity.

The pseudo-first order rate constants for the cleavage of (**1**) with 0.01 mol dm^{-3} CyD-N-2-N at pH 7.0 and 11.0 were 4.6×10^{-3} and 6.2×10^{-2} min^{-1}, respectively. The catalytic activity at pH 11.0 largely (more than 10^4 fold) surpasses the activity of the $[Co(en)_2(OH)(H_2O)]^{2+}$ complex (en = ethylenediamine), one of the most active catalysts hitherto studied for the cleavage of phosphodiesters.[4] The rate constant for CyD-N-2-N at pH 7.0 is close to the value (2.5×10^{-2} min^{-1}) of the cobalt(III) complex.

The products promptly leave the catalysts without the formation of any stable intermediates. In the cobalt(III) complex-catalysed cleavage of phosphodiesters, however, the products bound to the complex are released with difficulty.[4]

The present results show that the formation of catalytic sites of the artificial nucleases from only organic residues is possible if there is sufficient co-operation between the residues.

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Table 1. Rate constants k for the catalytic cleavage of (**1**) by the diamines at 50 °C.

	k/10^{-3} mol^{-1} dm^3 min^{-1}	
Diamine	Monocation	Neutral diamine
N–1–N	87	182
N–2–N	7.4	204
N–3–N	19	140
[NMe_3 (neutral form)]	[<0.1]	

References

1 (a) P. B. Dervan, *Science*, 1986, **232**, 464; J. K. Barton, *ibid.*, 1986, **233**, 727; D. S. Sigman, *Acc. Chem. Res.*, 1986, **19**, 180; L. A. Basile, A. L. Raphael, and J. K. Barton, *J. Am. Chem. Soc.*, 1987, **109**, 7550.

2 B. Barbier and A. Brack, *J. Am. Chem. Soc.*, 1988, **110**, 6880.

3 A. J. Kirby and W. P. Jencks, *J. Am. Chem. Soc.*, 1965, **87**, 3209.

4 J. Chin and X. Zou, *J. Am. Chem. Soc.*, 1988, **110**, 223; J. Chin, M. Banaszczyk, V. Jubian, and X. Zou, *ibid.*, 1989, **111**, 186; J. Chin and M. Banaszczyk, *ibid.*, 1989, **111**, 4103.