Efficient Cleavage of Adenylyl(3′-5′)adenosine by Triethylenetetraminecobalt(III)

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Cleavage of adenylyl(3′-5′)adenosine is 10^5 fold accelerated by [Co(triethylenetetramine)$(H_2O)_2$]$^{3+}$ (0.2 M) at pD 6, 50 °C.

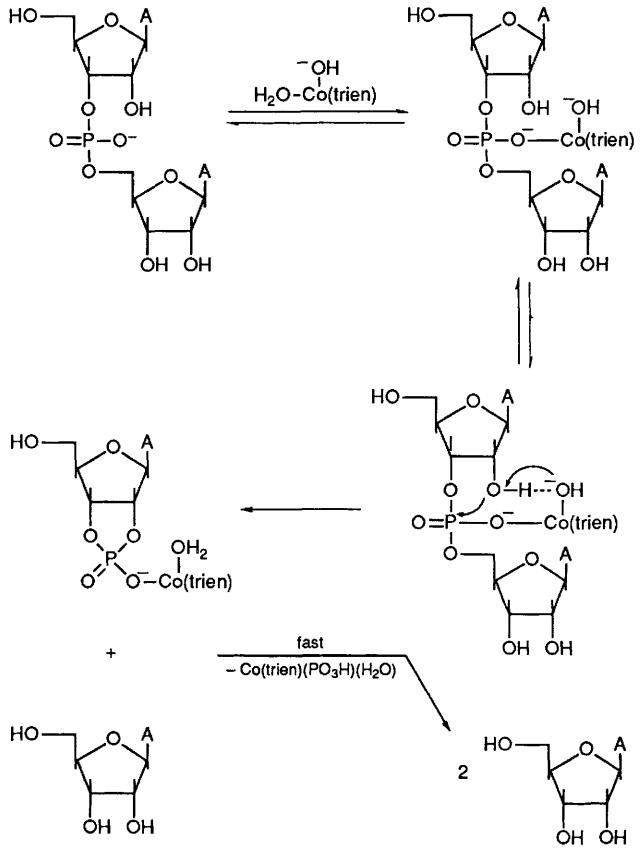
Recently artificial nucleases for the oxidative cleavage of nucleic acids, primarily at the ribose residues, have attracted much interest.[1] However, artificial systems involving fission of the phosphodiester linkage are scarce.[2] Catalytic residues showing large activities for the fission are required.[3—7]

This communication reports that [Co(trien)$(H_2O)_2$]$^{3+}$ (trien = triethylenetetramine) effectively catalyses the cleavage of adenylyl(3′-5′)adenosine (ApA) *via* fission of the phosphodiester linkage under mild conditions (pD 5—8; 50 °C).

Efficient cleavage of ApA to two adenosine molecules by [Co(trien)$(H_2O)_2$]$^{3+}$, prepared *in situ* by hydrolysis of *cis*-[Co(trien)Cl_2]Cl, is clearly evidenced by 500 MHz ^{1}H NMR spectroscopy. The signals for ApA (the C-1′–H protons of the ribose residues of adenosine on the 3′-side and the 5′-side at δ 5.86 and 5.98, respectively)[8] monotonically decrease, accompanied by the increase of the C-1′–H signal for free adenosine at δ 6.07 in a stoicheiometric proportion. The first-order rate constant at pD 6, 50 °C, increases almost linearly with increasing concentration of the complex, and is 5.2×10^{-5} min^{-1} at [complex]$_0$ = 0.2 M.

In the absence of the complex, however, the cleavage is too slow at pD 6 to be directly followed. The rate constant estimated from the pH–rate constant profile (a fairly straight line of slope 1.0 at pH 8—13 obtained by HPLC) is 3×10^{-10} min^{-1}. Thus acceleration by the complex (0.2 M) is 10^5 fold, decreasing the half-life of ApA from 4000 years to 9.3 days.

The facile cleavage of ApA to adenosines by the complex is



Scheme 1. Proposed mechanism for the $[Co(trien)(H_2O)_2]^{3+}$-catalysed cleavage of ApA.

further confirmed by HPLC; the rate of appearance of adenosine agrees reasonably with the rate constant evaluated by NMR spectroscopy. The pH–rate constant profile implies that hydroxide ion co-ordinated to the Co^{III} ion is responsible for the catalysis (pK_a values of the Co^{III}-co-ordinated water molecules are around 5.5 and 8).[7]

The Co^{III} complex exhibits no measurable catalysis for the cleavage of 2′-deoxyadenylyl(3′-5′)-2′-deoxyadenosine. The 2′-hydroxy residues in ApA are essential for the catalysis.

The proposed mechanism for the cleavage involves co-ordination of the phosphate of ApA to the Co^{III} complex, as depicted in Scheme 1. Then the hydroxide ion on the Co^{III} ion functions as a general base catalyst, promoting intramolecular nucleophilic attack of the 2′-hydroxy residue on the phosphorus atom. The resultant 2′,3′-cyclic phosphate of adenosine is promptly converted to the final product adenosine, catalysed by the complex, without accumulation in a detectable amount. This reaction scheme is supported by the efficient cleavage of adenosine 2′,3′-cyclic phosphate, obtained independently, to adenosine by the complex; the rate constant is more than 10^5 times as large as the value for the cleavage of ApA.

The present finding indicates that the Co^{III} complex is a potent candidate for the catalytic site of artificial ribonucleases.

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