

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

Yoichi Matsumoto and Makoto Komiyama*

Institute of Materials Science, University of Tsukuba, Tsukuba, Ibaraki 305, Japan

Cleavage of adenylyl(3'-5')adenosine is 10^5 fold accelerated by $[\text{Co}(\text{triethylenetetramine})(\text{H}_2\text{O})_2]^{3+}$ (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

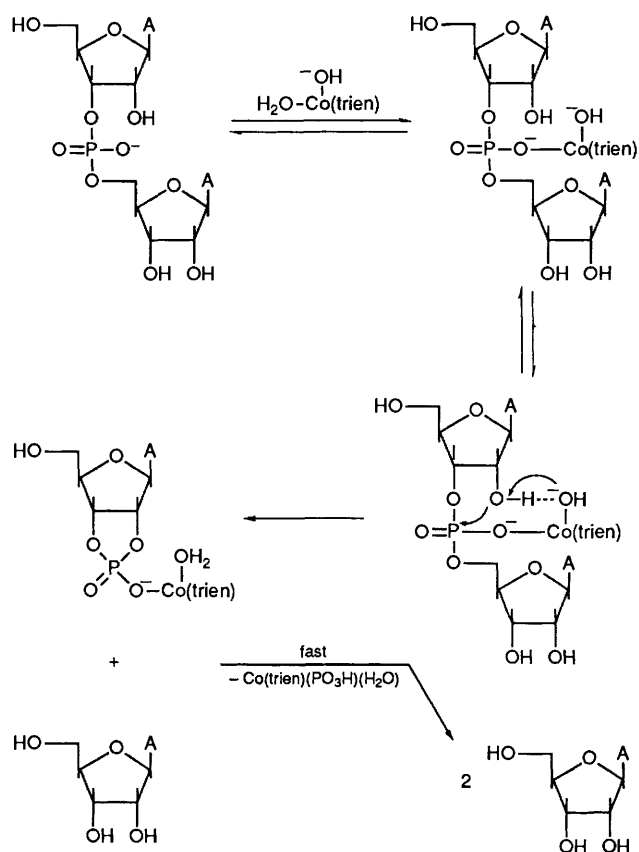
This communication reports that $[\text{Co}(\text{trien})(\text{H}_2\text{O})_2]^{3+}$ (trien = triethylenetetramine) effectively catalyses the cleavage of adenylyl(3'-5')adenosine (ApA) *via* fission of the phosphodiester linkage under mild conditions (pD 5-8; 50 °C).

Efficient cleavage of ApA to two adenosine molecules by $[\text{Co}(\text{trien})(\text{H}_2\text{O})_2]^{3+}$, prepared *in situ* by hydrolysis of *cis*- $[\text{Co}(\text{trien})\text{Cl}_2]\text{Cl}$, is clearly evidenced by 500 MHz ¹H 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 stoichiometric proportion. The first-order rate constant at pD 6, 50 °C, increases almost linearly with increasing concentration of the complex, and is $5.2 \times 10^{-5} \text{ min}^{-1}$ at $[\text{complex}]_0 = 0.2 \text{ M}$.

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 $3 \times 10^{-10} \text{ min}^{-1}$. 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 $[\text{Co}(\text{trien})(\text{H}_2\text{O})_2]^{3+}$ -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 ($\text{p}K_{\text{a}}$ 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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