

**SYNTHESIS AND CHARACTERIZATION OF A REACTIVE BINUCLEAR Co(III) COMPLEX.  
COOPERATIVE PROMOTION OF PHOSPHODIESTER HYDROLYSIS**

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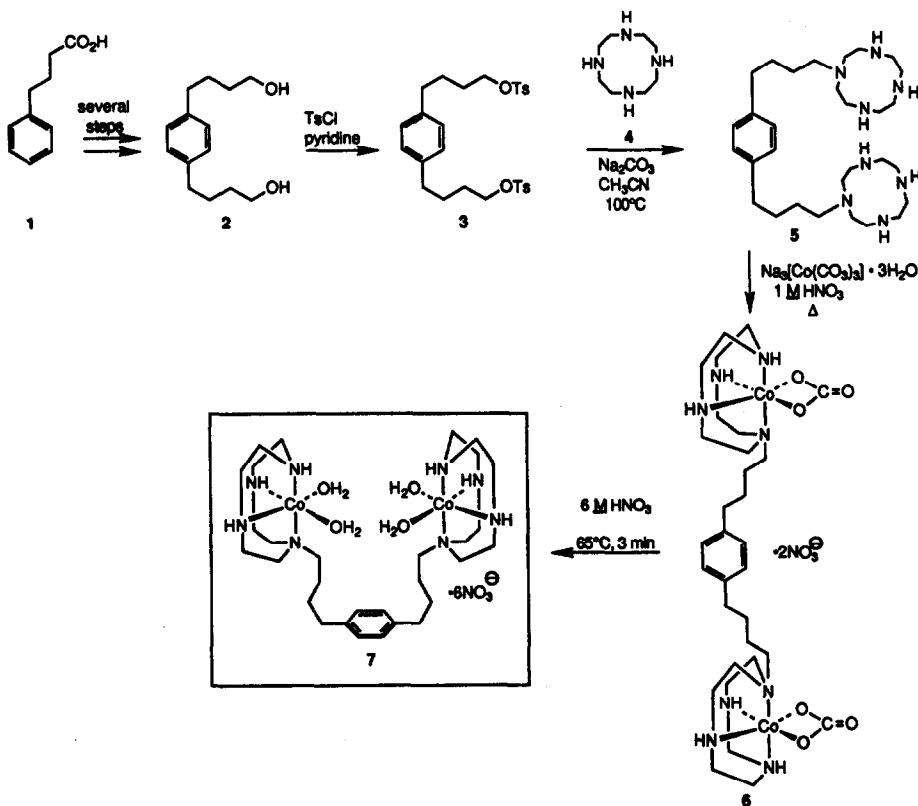
**Summary:** A binuclear Co(III) complex has been prepared that shows greater reactivity towards bis(p-nitrophenyl)phosphate than two equivalents of the parent mononuclear Co(III) complex.

The enzyme-like cleavage of phosphate esters under physiological conditions is of great interest due to the biochemical relevance of both naturally-occurring (e.g., DNA) and synthetic (e.g., nerve toxins) derivatives. The cleavage of DNA has been accomplished with specificity through the use of selectively delivered metal ion complexes.<sup>1</sup> However, most of the complexes used to date for this purpose cleave DNA via oxidative mechanisms that do not form religatable ends; this potential shortcoming has been addressed using both selectively delivered non-redox metals<sup>1f</sup> and RNase conjugates.<sup>2</sup> Several groups have worked to improve the use of completely synthetic, abiotic reagents and catalysts for phosphate ester hydrolysis.<sup>3</sup> While many metals provide accelerations in phosphate ester hydrolysis,<sup>4</sup> Co(III) complex cleavage of both phosphate esters and phosphoanhydrides has been most studied.<sup>5</sup> Recently, Chin has shown that structural variations on the Co(III) ligand can increase the reactivity of complexes towards phosphate ester cleavage;<sup>6</sup> tren, cyclen, and trpn complexes of Co(III) cleave bis(p-nitrophenyl)phosphate at 50°C with relative rates of ca.  $3 \times 10^7$ ,  $2 \times 10^9$ , and  $10^{10}$ -fold over that of the nonpromoted reaction.<sup>6d</sup>

However, the reaction of a single equivalent of Co(III) with phosphoanhydrides yields complexes stable towards cleavage;<sup>7</sup> reaction becomes fast only when additional equivalents of

Co(III) are added.<sup>5d-f</sup> More pertinently to phosphate esters, the 1:1 trpn-Co(III) complex with 5'-AMP is hydrolytically stable; addition of a second equivalent of Co(III) results in rapid, quantitative dephosphorylation to afford adenosine and the bidentate  $\mu^4$ -phosphato complex.<sup>6a</sup> While not catalytic, this method appears to have enormous potential for the efficient, hydrolytic cleavage of phosphate esters. Therefore, we were interested in learning whether or not a kinetic advantage would be obtained by a binuclear complex in which both metals are present in a single molecule. During the course of this work, a similar approach was reported by Breslow who showed a binuclear Zn(II) complex to be more effective toward the hydrolysis of p-nitrophenyl diphenyl phosphate than a reference mononuclear Zn(II) complex.<sup>8</sup>

The conversion of 4-phenylbutyric acid (1)<sup>9</sup> to 1,4-bis(hydroxybutyl)benzene (2) was carried out as described by Cram,<sup>10</sup> and ditosylate 3 (mp 61-63°C) was obtained by tosylation in pyridine. Condensation with 25 equivalents of cyclen (4) afforded the biscyclenyl derivative 5; excess cyclen, which is relatively expensive, was recovered in 80% yield by sublimation (95°C, 0.2 torr). Purification of 5, which is an oil, was



accomplished by selective extraction between  $\text{CHCl}_3$  and 10% aq.  $\text{Na}_2\text{CO}_3$  and drying and evaporation of the organic phase. Both  $^1\text{H}$  and  $^{13}\text{C}$  NMR confirmed the symmetry of 5.

Reaction of 5 with  $\text{Na}_3[\text{Co}(\text{CO}_3)_3] \cdot 3\text{H}_2\text{O}$  in 1 M  $\text{HNO}_3$  followed by washing with MeOH and acetone gave biscarbonato complex 6 in 87% as a pink solid,  $\lambda_{\text{max}}$  ( $\text{H}_2\text{O}$ ) 370 nm.  $^{13}\text{C}$  NMR of 6 reveals 12 alkyl, 2 aromatic, and 1 carbonyl carbon, consistent with a single symmetry plane reflecting one Co complex onto the other. Because the parent cyclen-Co(III) carbonato complex demonstrates only an equatorial (i.e., in the O-Co-O plane) and not an apical symmetry plane (the  $^{13}\text{C}$  NMR shows four lines, not one), alkylation of an apical nitrogen is indicated as shown in 6.

Direct hydrolysis of 6 to bis(diaquo) complex 7 was accomplished in 95% yield by heating in 6 M  $\text{HNO}_3$  at  $65^\circ\text{C}$  for 3 min. Evaporation to dryness, vacuum desiccation, and washing with ether provided 7, characterized by  $^1\text{H}$  and  $^{13}\text{C}$  NMR, UV, FAB MS [ $m/e$  1029 ( $\text{M}^+ - \text{NO}_3 - 1$ ), 771 ( $\text{M}^+ - 4\text{NO}_3 - 4\text{H}_2\text{O} - 1$ ), 642 ( $\text{M}^+ - 6\text{NO}_3 - 4\text{H}_2\text{O} - 6$ ), and CHN microanalysis (H low by 0.55%).

CPK models predict that 7 is sterically unable to form an intramolecular  $\mu$ -oxo dimer, which would lead to loss of reactivity towards phosphates; however, interaction through an intervening phosphate ligand appears possible. In fact, the  $^{31}\text{P}$  NMR ( $\text{D}_2\text{O}$ ) of  $\text{Na}_2\text{HPO}_4$  in the presence of one equivalent of 7 shows resonances at 17 ppm (v. broad), 28 ppm (broad), and 36-39 ppm (5 singlets) corresponding respectively to phosphate species bearing 2, 3, and 4 P-O-Co(III) bonds.<sup>6e,11</sup> While only conjecture, it is tempting to hypothesize that some or all of the tetracoordinated phosphate species are spirocyclic forms of "chelated" phosphate, made isomeric by nature of the relative cyclen-Co orientations. Distinguishing between chelated and "linear" forms of tetracoordinated phosphate remains a topic of interest.

More direct evidence for the cooperative interaction of intramolecular Co centers comes from the observed rates of complex-promoted bis(*p*-nitrophenyl)phosphate (BPNPP) hydrolysis. Uncatalyzed phosphodiester hydrolysis is extremely slow, with a  $k_{\text{obs}}$  at  $25^\circ\text{C}$  and pH 7 of about  $1 \times 10^{-12} \text{ s}^{-1}$ . Addition of the parent cyclen-Co(III) complex (2 mM) to a 50  $\mu\text{M}$  solution of BPNPP results in the loss of *p*-nitrophenol with  $k_{\text{obs}} = 2.2 \times 10^{-5} \text{ s}^{-1}$ , a roughly  $10^7$ -fold acceleration. However, addition of 1 mM 7 (overall 2 mM in Co) under the same conditions affords a hydrolysis rate 3.2-times faster still ( $k_{\text{obs}} = 7.0 \times 10^{-5} \text{ s}^{-1}$ ). We

attribute this modest, but real effect to the high effective concentration of a second Co complex once anation to the first has occurred. Of course, there are many degrees of freedom in 7 that could be removed, potentially leading to a greater intramolecular advantage; the present work provides a justification for the synthetic effort required to make more rigidly convergent binuclear dephosphorylation reagents or catalysts.

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