The interactions mentioned can not so far be expressed in quantitative ways nor can their contributions to equilibrium constants of macromolecular complex formation be written explicitly, therefore the way of describing polymeric complexing systems is through the use of averaged values.

Various examples of the interactions of metal ions with macromolecular chains of DNA and RNA, pH-dependence, conformational changeability and reversibility, as well as the possibility of modelling the macromolecules under study are given.

T19

A Rapid Kinetic Study of Divalent Metal Interactions with Flavin Coenzymes

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Research in these laboratories has focussed in recent years on the kinetics of divalent metal ion interactions with coenzymes. The principal kinetic tool has been temperature-jump relaxation spectroscopy. A large amount of kinetic information is now available for several nucleotides (*e.g.* AMP) and inorganic phosphates.

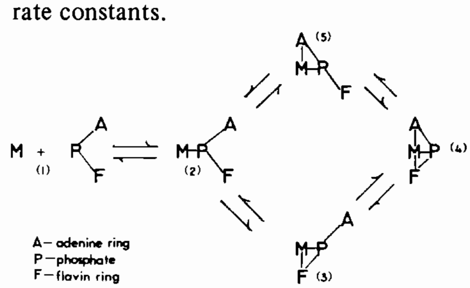
The purpose of this paper is to report the first rapid kinetic study of the mechanism of divalent metal ion interactions with the coenzymes flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD). The two compounds are structurally related to each other as well as to other coenzymes and phosphates that we have previously studied. FAD, for example, is structurally a combination of riboflavin phosphate and adenosine monophosphate (AMP). Ni(II) was chosen as the metal ion for these studies because of the large body of kinetic information that is already available for that ion. It can serve as a useful representative of divalent transition metal ion interactions with these and related coenzymes.

The Ni–FMN system. Two relaxation effects were observed in the kinetic experiments: one (τ_1) on the order of 0.2 msec, the other (τ_2) at about 2 msec. The detailed concentration and pH dependencies of τ_1 and τ_2 are quite similar to those for the relaxation times found in the Ni-ribose phosphate and Ni–AMP systems respectively. The mechanism consistent with these observations is a dual-pathway, back-bound complex mechanism, shown schematically as I:

$$\begin{array}{ccccc} 1 & & 2 & & 3 \\ \mathrm{Ni} + \mathrm{L} & \rightleftharpoons & \mathrm{NiL} & \rightleftharpoons & \mathrm{NiL'} \\ \Big\| K_A & & \Big\| K_B & & \Big\| K_C \\ \mathrm{Ni} + \mathrm{HL} & \rightleftharpoons & \mathrm{NiHL} & \rightleftharpoons & \mathrm{NiHL'} \\ 1' & & 2' & & 3' \end{array} \qquad \mathrm{(I)}$$

in which NiL is the phosphate-bound complex, NiL′ the phosphate + base bound complex.

The Ni–FAD System. FAD presents a number of different binding sites in the ionized phosphate bridge and the base nitrogens on both the adenine and isoalloxazine rings. The flexibility of this molecule facilitiates both individual and simultaneous ring interactions with the phosphate-bound metal. The Ni–FAD system is unique in that *four distinct* relaxation times, τ_1–τ_4, were found and characterized. The relaxation times ranged from 90 μsec to 20 msec and were found to be only slightly pH and concentration dependent. Based on the large body of prior data from our laboratory on simple nucleotide systems, we were able to associate specific relaxation times with reaction steps in scheme II and to determine the rate constants.



(II)

The mechanism shown as II quantitatively accounts for the number and behavior of all the relaxation steps.

In this scheme, F, P and A refer to the flavin, phosphate, and adenine moieties of the FAD molecule, respectively. The first step (1–2) involves bridging to the phosphate moiety only, followed by species involving interactions with the phosphate plus the flavin (5) or adenine moieties (3). The final complex (4) involves simultaneous interactions with all the components of the molecule.

T20

The Copper(II) Promoted Hydrolysis of Salicyl Phosphate (2-Carboxyphenyl Dihydrogen Phosphate)

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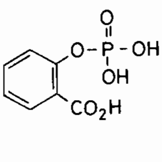
One of the striking observations of biological phosphate chemistry is that much of it appears to be subject to metal ion catalysis. However, the role of the metal ion in promoting the hydrolysis reactions of phosphate derivatives including phosphate esters has been the subject of considerable speculation.

Copper(II) ions have been observed to catalyse the hydrolysis of a number of phosphate monoesters including salicyl phosphate (I) [1, 2], 8-quinolyl phosphate (II) [3, 4] and 2-(4(5)-imidazoyl)phenyl phosphate (III) [5]. The catalytic effect observed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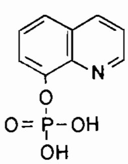
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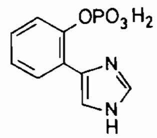
(III)

with salicyl phosphate and 8-quinolyl phosphate was apparently quite small (*ca.* 10 fold).

We have studied the copper(II) promoted hydrolysis of salicyl phosphate over a range of copper(II) concentrations at pH 4.72, 5.14 and 5.30 at 30 °C and I = 0.1 *M* (KNO_3). Copper(II) ions exert a very marked effect on the hydrolysis of the normally unreactive phosphate monoester dianion of salicyl phosphate (*ca.* 10^{10} rate acceleration). Previous work in this area had indicated only small rate accelerations, as comparisons were made between the metal ion promoted reaction and the *intramolecular general acid catalysed hydrolysis* of the phosphate monoester dianion.

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T21

Physico-Chemical Investigation of Nucleoside-Containing Pt(II) Triamines

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The discovery of the antitumor activity of *cis*-Pt($NH_3)_2Cl_2$ has aroused considerable interest in the study of Pt(II) complexes with nucleosides [1]. Most of the compounds investigated contain two nucleoside molecules and are of the nonelectrolyte or cation type [2–5].

We have synthesized and investigated isomeric Pt(II) triamines of composition $[Pt(NH_3)_2LCl]Cl$, where L = adenosine(ado), inosine(Ino), and *cis*-$[Pt(NH_3)_2L'Cl]Cl$, where L' = cytidine(Cyd).

The coordination formulae have been proved by the measurement of molecular conduction (Λ in aqueous solution = 100–110 (ohm^{-1} cm^2 mol^{-1}) and by long-wave IR spectroscopy ($\nu_{Pt\text{-}Cl}$ lies in the range 330–337 cm^{-1}). The hydrolysis constant K_h of the above triamines and $[Pt(NH_3)_3Cl]Cl$ has been determined potentiometrically with the use of Ag/AgCl and chloroselective electrodes.

$K_h \times 10^4$:

$[Pt(NH_3)_3Cl]Cl$ 2.3 *trans*-$[Pt(NH_3)_2InoCl]Cl$ 7.0

trans-$[Pt(NH_3)_2Clado]Cl$ 9.0 *cis*-$[Pt(NH_3)_2CydCl]Cl$ 20.6

cis-$[Pt(NH_3)_2adoCl]Cl$ 19.0

The substitution of NH_3 by a purine or pyrimidine molecule leads to an increase in K_h which is likely to be due to steric factors. The hydrolysis constant K_h is slightly affected by the nature of the nucleoside.

The geometric structure of the complexes affects K_h: *cis*-isomers are approximately 2–3 times less stable than *trans*-isomers. The lower stability of the Pt–Cl bond in the *cis*-triamine $[Pt(NH_3)_2 \cdot adoCl]Cl$ also follows from the comparison of $\nu_{Pt\text{-}Cl}$ in isomers: *cis*, 330 cm^{-1}; *trans*, 337 cm^{-1}.

The acidic properties of isomers $[Pt(NH_3)_2InoCl]Cl$ have been investigated by the method of potentiometric titration with an alkali in the presence of 0.3 *N* KCl (Fig. 1). Coordination leads to the enhancement of the acidic properties of inosine: pK_a of the

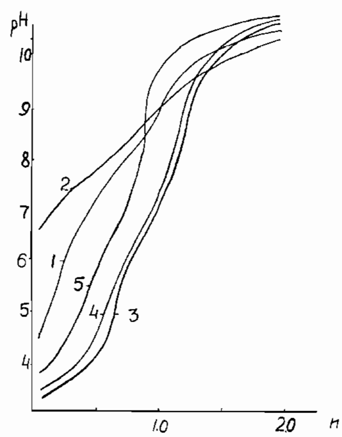

Fig. 1. pH values *versus* the number of added equivalents of OH-ions (n). $[Pt(NH_3)_2InoCl]Cl$: 1-*trans*, 2-*cis*; $[Pt(NH_3)_2InoH_2O] \cdot (NO_3)_2$: 3-*trans*, 4-*cis*; 5-*cis*$[Pt(NH_3)_2Ino](NO_3)_2$. Concentration of complexes = 1.10^{-3} *M*.