Catalysis of Isotopic Hydrogen Exchange in 1-Methylimidazole by Cr(III)

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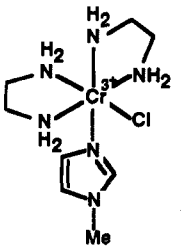
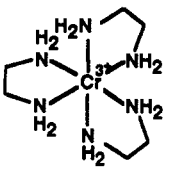
The ability of metal ions to bind to the nitrogen centers of imidazole and related heterocyclic portions of biomolecules, coupled with the wide occurrence of the imidazole nucleus in many biologically active molecules such as proteins, enzymes, vitamins, and nucleic acids, highlights studies of the relationship between the vital physiological roles of metal ions in living systems and the imidazole nucleus.[1,2] A useful handle for mechanistic studies is provided by the acidity of C(2)–H and C(8)–H in imidazole and in purines.[3,4] Thus a number of studies have been undertaken to evaluate metal ion effects on C(2/8)–H exchange in imidazoles,[5] histidine,[3b,6] purines,[6,7] and similar substrates as chemical probes of the biological action of metal ions. In general, the information that has resulted from these studies indicates that metal ions such as Pd(II), Pt(II), Cu(II), Ag(I), and CH_3-Hg^{II} are far less effective than H^+ in promoting C–H exchange in these molecules. Consequently, this behavior has been regarded as general.

In this communication, we report kinetic results obtained from the detritiation of the imidazole moiety of the *substitution-inert*[8] complex **1** which show conclusively that Cr(III) is a better catalyst than H^+ for this process. The study provides the first example of a more effective catalytic role by a metal ion relative to H^+. As well, the results show that the relatively inert C(4,5)–H in the free ligand[9] are significantly activated through coordination of the ligand to Cr(III), to the extent that tritium exchange in these positions occurs at convenient rates under the relatively mild conditions of the study.

cis-$[Cr(en)_2MeImCl]Cl_2$ {en = ethylenediamine, MeIm = 1-methylimidazole} (**1**) was prepared from *cis*-$[Cr(en)_2Cl_2]Cl\cdot H_2O$ and 1-methylimidazole and gave satisfactory analysis. Tritiation of the complex was done according to the method reported previously.[3] Detritiation of **1** in aqueous buffers under first-order conditions was followed at 35 °C by liquid scintillation counting technique. The initial counts (dpm) in these kinetic



1



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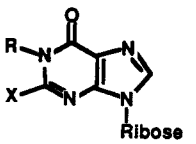

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a: R = CH_3, X = NH_2
b: R = H, X = H
c: R = CH_3, X = H

experiments were unusually high, and first-order plots, $\log(C_\infty - C_t)$ *vs* time, were curved, indicating the occurrence of two parallel first-order processes.[10] Separation of these rate processes according to the method of Frost and Pearson[11] afforded observed first-order rate constants k^A_{obs} and k^B_{obs} for the fast and slow exchange reactions, respectively. The corresponding second-order rate constants at 35 °C obtained from linear plots of k_{obs} *vs* $[OH^-]$ are $k^A = 6.0 \times 10^3\ M^{-1}\ s^{-1}$ and $k^B = 7.1 \times 10^2\ M^{-1}\ s^{-1}$. For direct comparison, detritiation rates for N–T exchange of **2** were also measured. No evidence for exchange of the -CH_2-protons of the ethylenediamine moiety was found under the conditions of the study. The second-order rate constant for the single (N–H) exchange process was calculated as $k = 5.0 \times 10^5\ M^{-1}\ s^{-1}$. It is expected that the rate of N–H exchange in the en moiety of **1** and **2** would be similar, since substitution of 1-methylimidazole and chlorine in **1** would have presumably moderate but compensating effects.[12]

Of the hydrons (L = H, D, or T) potentially capable of undergoing isotopic exchange in **1**, the most acidic is N–L of the ethylenediamine moiety. The unusually high counts (dpm) obtained in the initial assay of **1** (*vide supra*) constitute good evidence that N–T → N–H exchange had occurred by the time the first experimental point was acquired and is in accord with the *ca.* 10^2-fold difference in k^A for **1** and k for **2**, favoring **2**.

We assign the fast and slow processes in **1** to C(2)–T and C(4,5)–T exchange, respectively, based on the available evidence in the literature, both from deuteration[6,9] and detritiation[7] studies, that C(2)–H in imidazoles is much more acidic than the other ring hydrogens. It is not possible to specify which position, C(4) or C(5), is favored in the slow exchange, although inductive/field effects would indicate preference for C(4) over C(5).

An indirect comparison can be made of the effect of Cr(III) coordination in **1**, as opposed to protonation of 1-methylimidazole, on C(2)–T abstraction by OH^-. We have previously obtained[3a] the value $k = 1.0 \times 10^4\ M^{-1}\ s^{-1}$ at 85 °C for C(2)–T exchange in $MeImH^+$, which is close to the k^A value of $6.0 \times 10^3\ M^{-1}\ s^{-1}$ found in this study for exchange in **1** at 35 °C. From this qualitative comparison of rates alone, it is clear that Cr(III) activates C(2)–H exchange more effectively than the proton. A more quantitative estimate can be made if one uses the E_a value of 16.5 kcal mol^{-1} obtained by Jones[7c] for the detritiation of [8-^{3}H]-adenine, which appears to be the closest analogous process for which a literature E_a value has been recorded. Use of this E_a value with the $MeImH^+$ system affords a rate constant at 35 °C of $2.9 \times 10^2\ M^{-1}\ s^{-1}$. Comparing the effects of the proton and Cr(III) on the activation of MeIm, Cr(III) is found to be ca. 20 times more effective as catalyst for C(2)–H exchange than H^+. It is also significant to note that C(4,5)–H exchange, which is observed at 35 °C under Cr(III) catalysis, occurs only at very high temperatures in the presence of H^+.[9] We therefore have

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here a clear case of superior catalytic ability of Cr(III) over H^+ for isotopic exchange of the ring protons in the imidazole nucleus. Moreover, since for H^+ catalysis the rate ratio for C(2)–H exchange over C(4)–H exchange[9a] is 3.5×10^4, while for Cr(III) catalysis this ratio is only ca. 10, one sees a leveling effect exerted by Cr(III). This is in accord with Cr(III) exerting largely an inductive effect on the exchange process, in which C(2)–H abstraction is favored over C(4)–H but not overwhelmingly so. However, in H^+ catalysis, C(2)–H abstraction leads to a resonance-stabilized ylide intermediate, while in C(4)–H abstraction, only the inductive effect is operative.

In contrast to the present results for the effect of Cr(III), the results of Brodsky et al.[5c] indicate that Co(III)-bound 1-methylimidazole undergoes sluggish deuteration at C(2)–H, pH 12, at room temperature, with $t_{1/2} = 16$ h. These results extrapolate to reactivity ratios of *ca.* 2×10^{-5}:1:20 for catalysis of C(2)–H exchange by Co(III), H^+, and Cr(III), respectively.[13] The inhibitory role of Co(III) as compared to the catalytic effect of Cr(III) in this reaction, relative to H^+, is reminiscent of changes in enzyme activity consequent upon changing the identity of the metal ion at the enzyme active site while keeping metal ion size and electrical charge constant.

Literature data consistently show that metal ions are poorer catalysts than the proton in isotopic hydrogen exchange in imidazole-type compounds. Thus, Jones and co-workers[5a] have established the following order of catalytic effectiveness in tritium exchange in imidazole: $H^+ \gg CH_3Hg^{II} > Ni(II) \approx Zn(II) > Cu(II)$. Co(III)-bound imidazole is inert to exchange in D_2O at 44 °C and pH 1, 5, and 8.1.[5b] In other imidazole-containing systems, the following order is evident for the catalytic effects of the different species indicated, from the original data of the different workers: (i) 1-methylhistidine (detritiation),[3b,6] $H^+ \gg CH_3Hg^{II} \geq Pd(II)$; (ii) 1-methylguanosine,[7,14] **3a** (detritiation) $H^+ \gg Cu(II)$; (iii) inosine,[6] **3b** (deuteration) $H^+ \gg Pt(II)$; and (iv) 1-methylinosine,[7,14] **3c** (detritiation) $H^+ \gg Cu(II) \gg Ag(I)$.

The results in the literature for OH^--induced exchange of C(2)–H (imidazoles) and C(8)–H (purines), as presented above, show that exchange occurs much faster in the protonated ligand than in the metal-coordinated species. *Thus the present result for Cr(III) is apparently the first example of a superior catalytic role by a metal ion relative to the proton in C(2)–H/C(8)–H exchange.*

The mechanism of H^+-catalyzed isotopic exchange in imidazoles involves rate-determining proton abstraction by hydroxide ion on the protonated substrate. Proton abstraction at C(2) leads to a resonance-stabilized ylide intermediate, while the intermediate formed on abstraction at C(4) or C(5) is stabilized by an inductive effect mechanism. In metal ion-catalyzed isotopic exchange in imidazoles, proton abstraction at all ring positions [C(2), C(4), and C(5)] would be largely influenced by the magnitude of the effective nuclear charge on the coordinated N(3) atom, though C(2)–H has intrinsically greater acidity.

The following factors, among others, would conceivably determine the magnitude of the effective nuclear charge of the metal ion (M^{n+}) coordinated to N(3) of imidazoles: (a) extent of M^{n+}–N σ-bond polarization; (b) the electronic structure of M^{n+}; (c) the importance of π metal-to-ligand back-bonding; and (d) the magnitude of ligand field stabilization energies. Further work is in progress to bring additional evidence to bear on this problem.

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(13) The comparisons involve ^3H out of the ligand for $[Cr(en)_2(Cl)MeIm]^{2+}$ (**1**) in H_2O, and similarly for $MeImH^+$, but ^2H into the ligand for $[Co(en)_2(OH)MeIm]^{2+}$ in D_2O. If one makes assumptions of approximate values for the solvent isotope effect and the substrate isotope effect, then the rate effect disfavoring Co(III) becomes even larger by a factor of ca. 10. We thank a referee for drawing this to our attention.

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