The Formation of an Antitubercular Complex $[Fe(CN)_5(INH)]^{3-}$ through Mercury(II)-Catalyzed Ligand Substitution Reaction: A Kinetic and Mechanistic Study

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ABSTRACT: The kinetics and mechanism of the formation of an antitubercular complex $[Fe(CN)_5(INH)]^{3-}$ based on the substitution reaction between $K_4[Fe(CN)_6]$ and isoniazid (INH), i.e., isonicotinohydrazide, catalyzed by Hg^{2+} in aqueous medium was studied spectrophotometrically at 435 nm (the λ_{max} of the golden-yellow-colored complex $[Fe(CN)_5(INH)]^{3-}$) as a function of pH, ionic strength, temperature, and the concentration of the reactants and the catalyst. The replacement of coordinated CN^- in $[Fe(CN)_6]^{4-}$ was facilitated by incoming ligand INH under the optimized reaction conditions: pH 3.5 ± 0.02, temperature $= 30.0 \pm 0.1°C$, and ionic strength $I = 0.05$ M (KNO_3). The stoichiometry of the reaction and the stability constant of the complex ($[Fe(CN)_5(INH)]^{3-}$) have been established as 1:1 and 2.10×10^3 M, respectively. The rate of catalyzed reaction was found to be slow at low pH values, to increase with increasing pH, to attain a maximum value at 3.50 ± 0.02, and finally to decrease after pH > 3.5 due to less

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availability of H^+ ions needed to regenerate the catalytic species. The initial rates were evaluated for each variation from the absorbance versus time curves. The reaction was found to be pseudo-first order with respect to [INH] and first order with respect to $[Fe(CN)_6^{4-}]$ at lower concentration, whereas it was found to be fractional order at higher [INH] and $[Fe(CN)_6^{4-}]$. The ionic strength dependence study showed a negative salt effect on the rate of the reaction. Based on experimental results, a mechanism for the studied reaction is proposed. The rate equation derived from this mechanism explains all the experimental observations. The evaluated values of activation parameters for the catalyzed reaction suggest an interchange dissociative (I_d) mechanism. © 2012 Wiley Periodicals, Inc. Int J Chem Kinet 44: 398–406, 2012

INTRODUCTION

Substitution at pentacyano(ligand)ferrate(II), $[Fe(CN)_5L]^{(n-3)-}$, is slow due to the strongly bonded cyanide forcing the iron(II) to adopt the low spin t_{2g}^6 electronic configuration, with maximum crystal field stabilization. There is extensive information available about ligand substitution reactions of pentacyano(ligand)ferrate(II) complexes [1,2]. However, the subject is still given considerable attention because of its fundamental importance in analytical and synthetic chemistry. An extensive series of complexes, substituted with N-, O-, S-, and P-donor ligands L^{n+} have been synthesized from labile $[Fe(CN)_5H_2O]^{3-}$ ion [3–6]. The kinetic, thermodynamic, and volume of activation data from numerous mechanistic studies are consistent with a dissociative mechanism for ligand substitution reactions in these complexes [7,8]. Because of their high stabilities in dark [9], the hexacyanoferrate(II/III) is considered nontoxic [10], but undergo photolysis in acidic medium [11] when exposed to ultraviolet (UV) radiation, producing $[Fe(CN)_5H_2O]^{3-}$ and CN^- or HCN. The primary slow reversible thermal aquation of $[Fe(CN)_6]^{4-}$ produces $[Fe(CN)_5H_2O]^{3-}$, which reacts with pyridine-4-carboxylic acid hydrazide or isonicotinic acid, commercially known as isoniazid (INH), yielding substituted antitubercular complex $[Fe(CN)_5(INH)]^{3-}$ through the following reactions:

$$[Fe(CN)_6]^{4-} + H_2O \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} [Fe(CN)_5H_2O]^{3-} + CN^-(\text{slow})$$

$$[Fe(CN)_5H_2O]^{3-} + INH \rightarrow [Fe(CN)_5(INH)]^{3-} + H_2O$$

$$CN^- + H_2O \rightleftarrows HCN + OH^-$$

The INH, an antitubercular drug, was first reported to be effective in the treatment of tuberculosis [12,13] but strains of *Mycobacterium tuberculosis*, a causative agent of disease resistant to INH, were also reported shortly after its introduction [14]. The mutations in drug target genes were held responsible for INH resistance [15–17]. The INH is a prodrug, and it is activated by mycobacterial catalase–peroxidase enzyme (CPE) in the presence of manganese ions, nicotinamide adenine dinucleotide (NADH), and oxygen [18–20]. The recent results have suggested that the complex, $[Fe(CN)_5(INH)]^{3-}$, requires no activation by CPE in the absence of NADH, and probably its mechanism of action involves interaction with NADH binding site of enzyme [21,22]. This inorganic analogue may represent a new class of leading antitubercular agents aiming at the inhibition of a validated target without activation of CPE in the absence of NADH [21,22].

However, such reactions involving exchange of the coordinated cyanide in least labile $[Fe(CN)_6]^{4-}$ by few nitrogen heterocyclic ligands [23–25] or $[Ru(CN)_6]^{4-}$ ion with two naphthalene substituted ligands, viz, nitroso-R-salt and α-nitroso-β-naphthol [26], are found to be very slow and it can be homogeneously catalyzed by some metal ions such as Ag^+ [23] and Hg^{2+} [24,25]. Such catalyzed reactions provided fundamental importance in the view of analytical chemistry for trace determination of metal ions in complex materials [24–28]. In such reactions, the catalyst Hg^{2+} abstracts a CN^--forming $HgCN^+$, which dissociates in acidic medium to regenerate the catalyst [24,25,29]. Thus, coordination of INH to transition metal ions in general and Fe(II) in particular is of special interest because of efficiency of this ligand to form a complex, $[Fe(CN)_5(INH)]^{3-}$. We have further been interested in studying the catalytic chemical and electrochemical wet oxidation of phenol using new copper(II) tetraazamacrocycle complexes under homogeneous conditions [30]. Therefore, in continuation of our interest in studying the catalytic reactions [23–28,30], in the present work we have considered it worthwhile to investigate the kinetics and mechanism of the Hg^{2+}-catalyzed antitubercular complex $[Fe(CN)_5(INH)]^{3-}$ formation through a ligand substitution reaction between $[Fe(CN)_6]^{4-}$ and INH.

EXPERIMENTAL

Reagents

All solutions were prepared using double distilled water throughout the study. All the reagents used were of analytical grade. $K_4[Fe(CN)_6]\cdot 3H_2O$ (Merck, Darmstadt, Germany), $HgCl_2$ (Glaxo Laboratories, Lucknow, India), and INH (BDH, Poole, UK) were used in this study. The stock solutions (1.0×10^{-2} M) of each of $K_4[Fe(CN)_6]\cdot 3H_2O$ and INH were prepared by their accurate weighing. $K_4[Fe(CN)_6]\cdot 3H_2O$ solution was stored in dark amber-colored bottles to avoid photodecomposition. The appropriate dilutions were made from these solutions immediately before use. Dilute solutions of $HgCl_2$ of required concentrations were prepared daily to prevent loss due to its adsorption on glass surface. The INH is highly soluble in water, and solution is quite stable and can be used for months. Buffer solutions were prepared by adding KCl to HCl or potassium hydrogen phthalate (Qualigens Fine Chemicals, India) and HCl/NaOH, as reported in the literature [31]. The standard BDH buffers were used to standardize the pH meter before use. KNO_3(Merck, Darmstadt, Germany) was used to maintain ionic strength in the reaction medium.

Instrumentation

A Shimadzu double-beam UV-240 spectrophotometer equipped with a circulatory arrangement of water for thermostating the cell compartment was used to monitor the progress of the reaction at $\lambda = 435$ nm, as well as repetitive spectral scan of the reaction mixture. The pH measurements were made on a digital CL46 Toshniwal (India) pH meter. A self-designed thermostat was used to maintain the temperature of all the reagents and the reaction mixture.

Product Characterization, Stoichiometry, and Kinetic Measurements

The reported antitubercular complex, formed between INH and $[Fe(CN)_6]^{4-}$, was synthesized by a direct reaction of $Na_3[Fe(CN)_5NH_3]\cdot 3H_2O$ with INH in aqueous solution at room temperature by adopting the procedure reported for cyanoferrate complexes [32]. The well-known complex has been fully characterized as $Na_3[Fe(CN)_5(INH)]\cdot 4H_2O$ using elemental analysis, nuclear magnetic resonance, UV–visible, Mössbauer spectroscopy, and cyclic voltametric studies [33]. The UV–visible spectrum of the complex $Na_3[Fe(CN)_5(INH)]\cdot 4H_2O$ in aqueous solution exhibited a peak at 436 nm ($\varepsilon = 4.0 \times 10^3$ M^{-1} cm^{-1}) [33] assigned to the metal to ligand charge-transfer

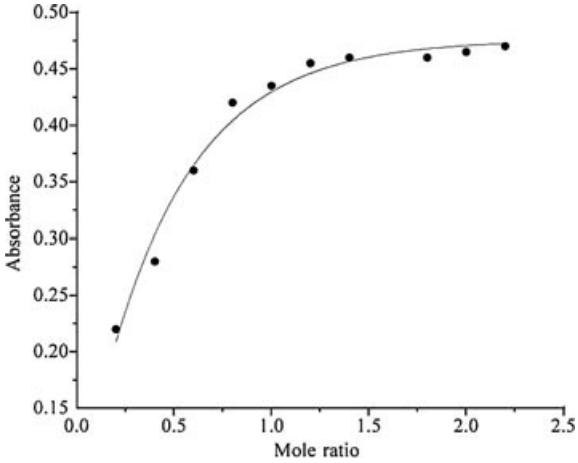
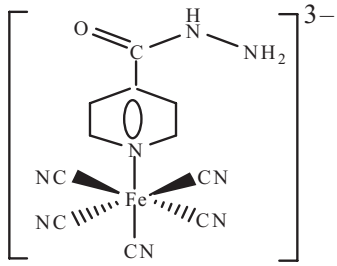


Figure 1 Determination of stoichiometry of the reaction by the mole ratio method at 2.5×10^{-4} M [INH], 2.5×10^{-6} M [Hg^{2+}], pH 3.5 ± 0.02, $I = 0.05$ M (KNO_3), and temperature $= 30.0 \pm 0.1$°C.

transition, Fe(II)d$\pi \rightarrow$ pπINH. The structure of the complex anion, $[Fe(CN)_5(INH)]^{3-}$ (see below), shows that INH coordinates through the ring nitrogen. The mole ratio study conducted on the complex formed during the course of the Hg^{2+}-catalyzed reaction between $[Fe(CN)_6]^{4-}$ and INH also revealed that the composition of the final reaction product is 1:1, i.e., $[Fe(CN)_5(INH)]^{3-}$ (Fig. 1). Hence, $[Fe(CN)_5(INH)]^{3-}$ is identified as the final reaction product during the course of the reaction between $[Fe(CN)_6]^{4-}$ and INH.



All reactant solutions were equilibrated at 30°C by keeping them for 30 min in a self-designed thermostat prior to the start of the reaction to maintain the desired temperature within the accuracy of ±0.1°C. The reactants (2.0 mL each) were mixed in a 50-mL Borosil measuring flask in the sequence: INH, buffer, and $HgCl_2$. The reaction was finally initiated by adding 2 mL of $K_4[Fe(CN)_6]\cdot 3H_2O$ solution to the reaction mixture. The reaction mixture was properly shaken to mix the contents and transferred quickly into a 10-mm cuvette in the temperature-controlled cell compartment of the spectrophotometer. The progress of reaction was subsequently followed by monitoring the increase in

absorbance at 435 nm, the λ_{max} of the golden yellow antitubercular complex $[Fe(CN)_5(INH)]^{3-}$ formed during the reaction. No absorbance correction was applied at the above wavelength because only this complex absorbs strongly and the other reactants, viz, $HgCl_2$, $K_4[Fe(CN)_6]$, and INH have no absorption. The initial rates (V_i) were calculated from absorbance values recorded at 435 nm and used to find the reaction order dependences in [INH], $[Fe(CN)_6^{4-}]$, and $[Hg^{2+}]$. The fixed time procedure was used to find the dependence of $[Hg^{2+}]$ and pH on the reaction rate. The effect of pH on the reaction rate was studied using potassium hydrogen phthalate–hydrochloric acid buffer to avoid any complication in the reaction system. No absorbance change at λ_{max} of INH was noticed on adding Hg^{2+} to its solution, which rules out any complexation between the two species.

RESULTS

Mercury(II)-Catalyzed Formation of Antitubercular Complex $[Fe(CN)_5(INH)]^{3-}$

The catalyzed ligand substitution reaction between $[Fe(CN)_6]^{4-}$ and INH has been investigated under various conditions by changing a desired experimental variable in turn while keeping all other variables constant. A detailed study of the kinetics and mechanism of this reaction is reported in this paper. The dependence of initial rates on different reaction variables are reported below and discussed at length separately.

Effect of pH on the Reaction Rate

The effect of pH on the initial rate of the reaction between $[Fe(CN)_6]^{4-}$ and INH catalyzed by Hg^{2+} ions was investigated using a fixed time procedure in the pH range 1.5–8.0 while keeping all other reaction variables constant to select a pH value corresponding to the optimum rate of reaction. In the acidic medium, the pH up to 6 was varied using KCl/HCl and potassium hydrogen phthalate/NaOH or HCl buffers. The plot of the absorbance A_t (measured after 5, 7, and 10 min of mixing the reagents) versus the pH of the reaction medium is shown in Fig. 2. It is evident from this plot that the rate is slow at low pH values, attains a maximum value at 3.50 ± 0.02, and subsequently falls again. The decrease in the rate at still higher pH values may be due to the deficiency of protons required to reproduce the catalytic species (vide infra). The lower rate of the reaction at low pH values is attributed to the formation of various protonated species of $K_4[Fe(CN)_6]$ having low reactivity [34]. The protonation consequently decreases the concentrations of free INH [35] as well as $[Fe(CN)_6]^{4-}$ needed for the reaction [23–25]. Thus, pH 3.5 was chosen for further experiment.

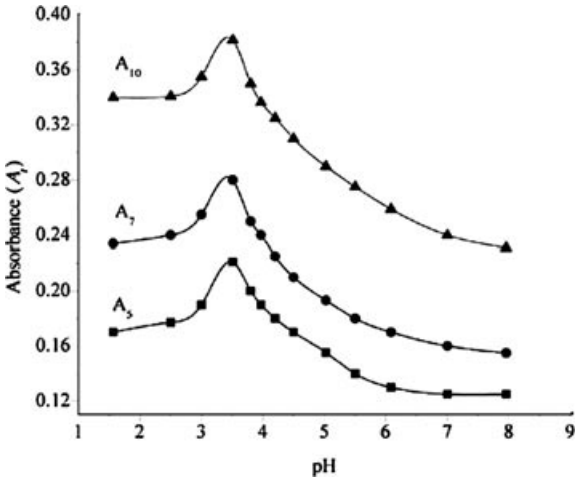


Figure 2 Effect of pH on Hg(II)-catalyzed substitution of CN^- in hexacyanoferrate(II) by INH at 1.25×10^{-2} M $[Fe(CN)_6^{4-}]$, 2.5×10^{-4} M [INH], 2.5×10^{-6} M $[Hg^{2+}]$, $I = 0.1$ M (KNO_3), and temperature $= 30.0 \pm 0.1$°C.

Effect of [INH] on the Reaction Rate

The initial rates were determined as a function of [INH] by varying its concentration from 2.5×10^{-5} to 4.5×10^{-4} M keeping the concentrations and other reaction variables fixed at 2.5×10^{-6} M $[Hg^{2+}]$, 2.5×10^{-3} M $[Fe(CN)_6^{4-}]$, pH 3.50 ± 0.02, $I = 0.05$ M (KNO_3), and temperature 30.0 ± 0.1°C. The plot of the initial rate (V_i) versus [INH] is shown in Fig. 3, which clearly indicates that the initial rate increases, showing

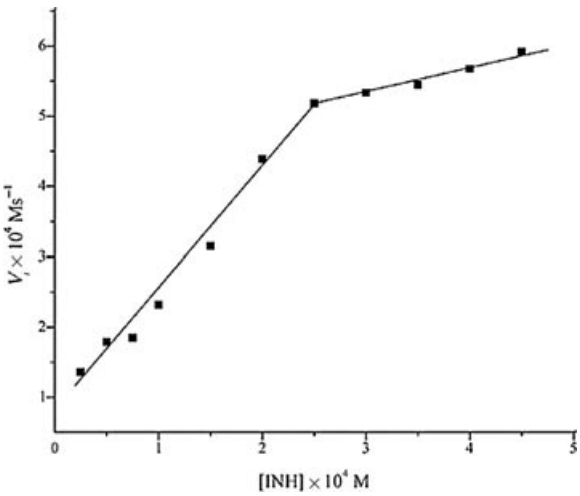


Figure 3 Effect of the INH concentration on the initial reaction rate at 2.5×10^{-2} M $[Fe(CN)_6^{4-}]$, 2.5×10^{-6} M $[Hg^{2+}]$, pH 3.5 ± 0.02, $I = 0.05$ M (KNO_3), and temperature $= 30.0 \pm 0.1$°C.

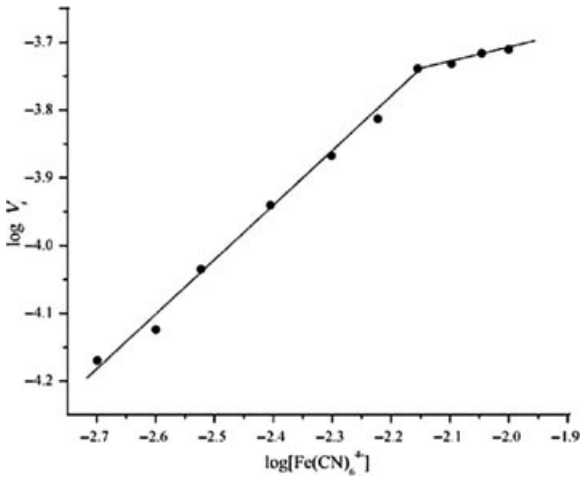


Figure 4 Dependence of the initial reaction rate on the concentration of $[Fe(CN)_6]^{4-}$ at 2.5×10^{-4} M [INH], $[Hg^{2+}] = 2.5 \times 10^{-6}$ M, pH 3.5 ± 0.02, $I = 0.05$ M (KNO_3), and temperature $= 30.0 \pm 0.1$°C.

a linear dependence up to 2.5×10^{-4} M [INH], and finally leveling off to almost a constant value at higher concentrations.

Effect of $[Fe(CN)_6^{4-}]$ on the Reaction Rate

The initial rates were evaluated as a function of $[Fe(CN)_6^{4-}]$ by changing its concentration from 1.0×10^{-4} to 1.0×10^{-2} M while keeping other experimental variables fixed at optimum values. The plot of log(initial rate) versus log$[Fe(CN)_6^{4-}]$ (Fig. 4) clearly indicates that the order of the reaction with respect to $[Fe(CN)_6^{4-}]$ is first order at its low concentration and a low fractional order at higher concentrations, but certainly not tending toward zeroth order. It has been demonstrated through an independent experiment that at higher $[CN^-]$ the rate of the forward reaction is inversely proportional to its concentrations.

Effect of Temperature on the Reaction Rate

The rate of Hg^{2+}-catalyzed ligand exchange between $[Fe(CN)_6]^{4-}$ and INH was studied as a function of temperature range 25–40°C keeping all other reaction variables fixed at an optimum value. Higher temperature was avoided due to the possibility of decomposition of the golden yellow complex, $[Fe(CN)_5(INH)]^{3-}$. The Arrhenius equation was used to determine the activation energy (E_a) for the catalyzed reaction. The energy of activation has been determined from the slope of the plot of $\log(V_i)$ versus $1/T$. The other activation parameters, viz, enthalpy of activation ($\Delta H^{\neq}$) and entropy of activation ($\Delta S^{\neq}$) have also been evaluated using Eyring's equation, $\ln(\text{initial rate}/T) = \ln(k_B/h \cdot e^{\Delta S^{\neq}/R}) - \Delta H^{\neq}/RT$, by plotting $\ln(V_i/T)$ versus $1/T$, which yields a straight line.

The values of activation parameters are found to be $E_a = 51.58$ kJ mol^{-1}, $\Delta H^{\neq} = 49.14$ kJ mol^{-1}, and $\Delta S^{\neq} = -144.73$ J K^{-1} mol^{-1}. The entropy of activation is highly negative in comparison to the corresponding positive values on pentacyano(ligand)ferrate(II) [34]. This highly negative value of entropy of activation points toward an enhanced degree of a bond-breaking (S_N1 character or dissociative character) in the activated complex of $[Fe(CN)_5L]^{(3-n)-}$ in comparison to corresponding $[Fe(CN)_6]^{4-}$ counterpart.

Effect of Ionic Strength (I) on the Reaction Rate

The effect of ionic strength on the initial rate of the reaction was studied employing KNO_3 for maintaining ionic strength in the 0.02–0.3 M range. Ionic strength >0.3 M was not studied due to the limited solubility of KNO_3. The results indicated that the initial rate decreases with increasing ionic strength of the reaction medium, confirming a negative salt effect on the rate of the reaction. The initial rate of the reaction was found to decrease drastically when KCl was used to maintain the ionic strength of reaction medium. Such reduction in the initial rate with increasing concentration of KCl, or in other words $[Cl^-]$, may be attributed to the formation of weakly ionized species of catalyst, viz, $HgCl_3^-$ and $HgCl_4^{2-}$ species, which decrease the effective concentration of catalytic species Hg^{2+} (or $HgCl^+$) in the reaction system.

Effect of $[Hg^{2+}]$ on the Initial Rate

The catalytic action of the mercury(II) concentration on the reaction rate at pH 3.5 was studied to check the linearity between the reaction rate and the $[Hg^{2+}]$ range for analytical application due to its catalytic effect, and also to gain an information into the changing role of mercury as a function of its concentration. The $[Hg^{2+}]$ was varied from 1.0×10^{-8} to 6.0×10^{-4} M keeping the concentrations of $[Fe(CN)_6]^{4-}$ and INH fixed at an optimum value of pH 3.5 ± 0.02, $I = 0.05$ M (KNO_3), and temperature $= 30.0 \pm 0.1$°C. The plot of the absorbance values A_t ($t = 2$, 5, and 7 min) as a function of $[Hg^{2+}]$ is shown in Fig. 5.

It is interesting to note that the catalytic activity of mercury increases, though not linearly, till the concentration of Hg^{2+} ions is approximately equal to the concentration of $[Fe(CN)_6]^{4-}$. The value of the intercept obtained by extrapolation of the initial linear portion of the curve (Fig. 5) corresponds to the rate due to uncatalyzed path. The observed decline in the rate at a

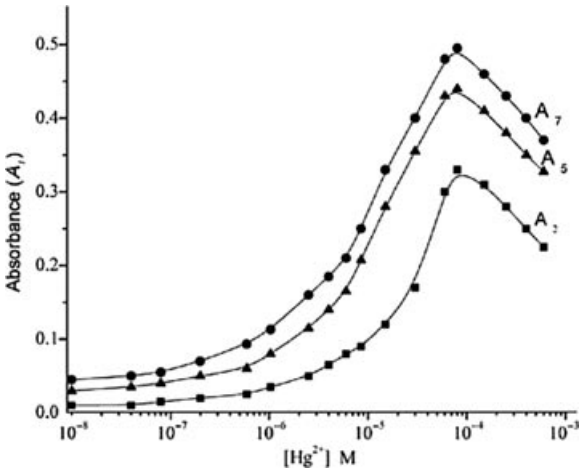


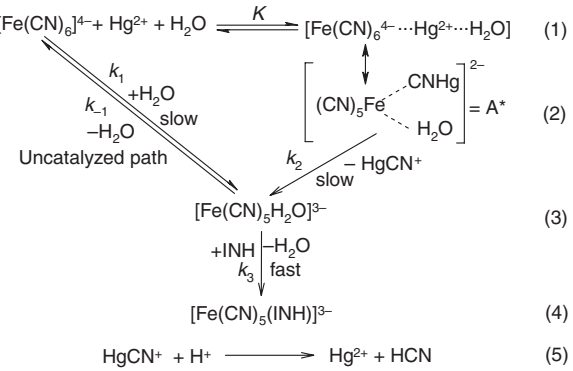
Figure 5 Effect of variation of $[Hg^{2+}]$ on the initial rate of the catalyzed ligand exchange reaction between hexacyanoferrate(II) and INH at 2.5×10^{-3} M $[Fe(CN)_6^{4-}]$, 2.5×10^{-4} M [INH], pH 3.5 ± 0.02, $I = 0.05$ M (KNO_3), and temperature $= 30.0 \pm 0.1$°C.

higher concentration of mercury may be due to formation of a binuclear adduct $[Fe(CN)_6^{4-} \cdot HgCl_2]$, which would decrease the rate of the forward reaction.

It has been observed through an independent experiment conducted at low $[Fe(CN)_6^{4-}]$ that mixing of $[Fe(CN)_6]^{4-}$ and $HgCl_2$ in a 1:2 molar ratio leads to the formation of white precipitate, which quickly turned into blue color. This observation further strengthens the idea of adduct/binuclear complex formations between $K_4[Fe(CN)_6]$ and $HgCl_2$ in 1:1 stoichiometric amount in solution. The existence of a similar complex formation between $[Fe(CN)_6]^{4-}$ and $Hg(CN)_2$ has been postulated by Beck [36].

DISCUSSION

The detailed experimental observation on the Hg(II)-catalyzed reaction between $[Fe(CN)_6]^{4-}$ and INH enabled us to propose the following mechanistic scheme through Eq. (1)–(5):



The rate expression for the formation of the complex $[Fe(CN)_5(INH)]^{3-}$ can be given as follows:

$$d[Fe(CN)_5(INH)^{3-}]/dt = k_3[Fe(CN)_5H_2O^{3-}][INH] \quad (6)$$

There are two possible paths for the formation of the intermediate $[Fe(CN)_5H_2O]^{3-}$, the catalyzed, as well as the uncatalyzed paths. The rate of the uncatalyzed path is dependent on the concentration of INH ($<2.5 \times 10^{-4}$ M), and this dependence varies with the concentration under certain conditions. The rate for the uncatalyzed path can be given by the following equation:

$$\text{Uncatalyzed rate} = k'\left[Fe(CN)_6^{4-}\right] \quad (7)$$

where k' is a composite rate constant involving some concentration terms and rate constants. If the slow decomposition of deactivated complex in the proposed mechanism (Eq. (2)) corresponds to the slowest or rate-determining step, the overall rate of the reaction due to both uncatalyzed as well as catalyzed reactions in a nonrate limiting concentration can be given through the following rate equation (8):

$$\begin{aligned} \text{Rate} &= d[Fe(CN)_5(INH)^{3-}]/dt \\ &= k'\left[Fe(CN)_6^{4-}\right] + k_2[A^*] \end{aligned} \quad (8)$$

In Eq. (8), the first and second terms correspond to the rate due to uncatalyzed and catalyzed reactions, respectively. Since

$$\begin{aligned} [Hg^{2+}]_T &= [Hg^{2+}]_{Free} \\ &+ \left[\text{Mercury adduct with } Fe(CN)_6^{4-}\right] \end{aligned} \quad (9)$$

Now on the basis of the proposed mechanistic scheme, Eq. (9) can be easily transformed to Eq. (10)

$$\begin{aligned} [Hg^{2+}]_{Total} &= [Hg^{2+}]_{Free} \\ &+ K\left[Fe(CN)_6^{4-}\right][Hg^{2+}]_{Free}[H_2O] \\ [Hg^{2+}]_{Free} &= [Hg^{2+}]_{Total}/\left(1+K\left[Fe(CN)_6^{4-}\right][H_2O]\right) \end{aligned} \quad (10)$$

In Eq. (10), K refers to the association constant between catalyst, water, and $[Fe(CN)_6]^{4-}$. The second term in Eq. (14) can be further expressed through Eq. (11)

$$k_2[A^*] = k_2K\left[Fe(CN)_6^{4-}\right][Hg^{2+}]_{Free}[H_2O] \quad (11)$$

Now putting the value of $[Hg^{2+}]_{free}$ from Eq. (10) into Eq. (11) yields Eq. (12):

$$k_2[A^*] = k_2K\left[Fe(CN)_6^{4-}\right][Hg^{2+}]_{Total}[H_2O]/ \left(1 + K\left[Fe(CN)_6^{4-}\right][H_2O]\right) \quad (12)$$

Now the rate of the overall reaction, both uncatalyzed and catalyzed, in the presence of nonrate limiting amounts of INH, i.e., Eq. (8), can be finally transformed to Eq. (13)

$$\text{Rate} = k'\left[Fe(CN)_6^{4-}\right] + k_2K\left[Fe(CN)_6^{4-}\right] \times [Hg^{2+}]_{Total}[H_2O]/(1 + K\left[Fe(CN)_6^{4}\right][H_2O]) \quad (13)$$

In Eq. (13), the first term corresponds to the rate due to the uncatalyzed reaction and second term corresponds to the rate due to the catalyzed reaction. The second term demonstrates the tendency toward leveling off in the rate at the higher concentration of $[Fe(CN)_6]^{4-}$, exhibiting a variable-order dependence in $[Fe(CN)_6]^{4-}$ in the presence of a small concentration of mercury. Equilibrium is supposed to lie on right-hand side of Eq. (1), and the value of K to be >1. When the concentration of $[Fe(CN)_6]^{4-}$ is smaller, i.e., $[Fe(CN)_6^{4-}] \ll 1$ and as the water concentration is in large excess, then Eq. (13) reduces to Eq. (14)

$$\text{Rate} = k'\left[Fe(CN)_6^{4-}\right] + k_2'K\left[Fe(CN)_6^{4-}\right][Hg^{2+}] \quad (14)$$

Equation (14) gives the observed rate constant as $k_{obs} = k' + k_2'[Hg^{2+}]$, where $k_2' = k_2[H_2O]$. However, at the higher concentration of $[Fe(CN)_6]^{4-}$, $K[Fe(CN)_6^{4-}] \gg 1$, Eq. (13) can be written as Eq. (15)

$$\text{Rate} = k'\left[Fe(CN)_6^{4-}\right] + k_2'[Hg^{2+}] \quad (15)$$

Thus, the rate constants k' and k_2' were evaluated from the intercept and slope, respectively, of a plot of the initial rate versus $[Hg^{2+}]$ in the presence of higher $[Fe(CN)_6^{4-}]$ (Eq. (15) and Fig. 6). The values of k' and k_2' obtained from the above plot at $I = 0.05$ M (KNO_3), temperature = 30.0 ± 0.1°C, and pH 3.5 ± 0.02 are given as 6.80×10^{-4} s^{-1} and 3.75 s^{-1}, respectively. The values of these rate constants were substituted in Eq. (14) to obtain the value of K corresponding to the various concentrations of Hg^{2+} at low values of $[Fe(CN)_6^{4-}]$. The average calculated values of K using Eq. (14) at varied $[Hg^{2+}]$ were found to be 241.4 ± 1.8. The average value of log K was found to be quite close to that calculated by Beck [36]. The value of the rate constant, $k_2 = 3.75$ s^{-1}, calculated at high $[Fe(CN)_6^{4-}]$ is shown to be valid at lower $[Fe(CN)_6^{4-}]$, where $K[Fe(CN)_6^{4-}] \ll 1$. k_2 calculated by a plot of rate versus $[Hg^{2+}]$ using Eq. (14) was found to be 3.78 s^{-1} and is in good agreement with $k_2 = 3.75$ s^{-1}.

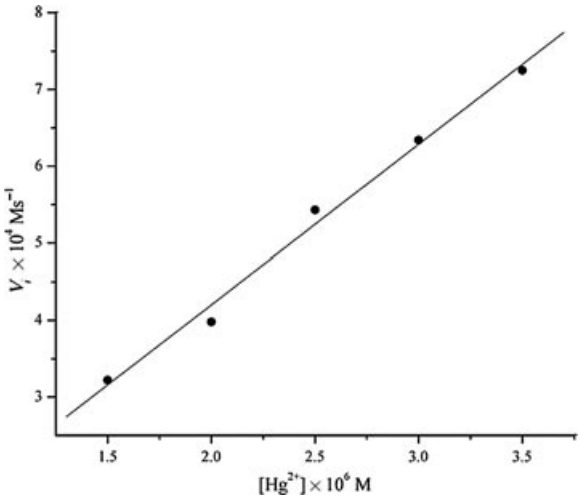


Figure 6 Effect of $[Hg^{2+}]$ on the initial rate at high $[Fe(CN)_6^{4-}]$ under condition: 5.0×10^{-3} M $[Fe(CN)_6^{4-}]$, 2.5×10^{-4} M [INH], pH 3.5 ± 0.02, $I = 0.05$ M (KNO_3), and temperature = 30.0 ± 0.1°C.

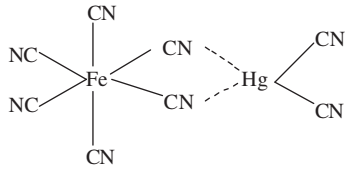
The rapid increase in the rate at $[Hg^{2+}] > 1.0 \times 10^{-5}$ M (Fig. 5) may be explained by considering the ionic character of the Hg–Cl bond in Hg_2Cl_2 along with the following equilibria:

$$Hg^{2+} + Cl^- \overset{K_1}{\rightleftharpoons} HgCl^+$$

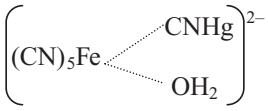
$$HgCl^+ + Cl^- \overset{K_1}{\rightleftharpoons} HgCl_2$$

On the basis of the species distribution ($HgCl_2$, $HgCl^+$, and Hg^{2+}), present in aqueous solution of $HgCl_2$, as a function of log[$HgCl_2$] [23–25], it has been found that at $[HgCl_2] > 10^{-6}$ M, the $HgCl^+$ concentration is considerable and at still higher concentration $HgCl_2$ is mainly present. Therefore, a loose adduct is formed with $HgCl^+$ in comparison to adduct formed with Hg^{2+} [37,38]. The adduct formed with $HgCl^+$and $[Fe(CN)_6]^{4-}$ decomposes faster than the adduct formed between $[Fe(CN)_6]^{4-}$ and Hg^{2+} and hence the rate is enhanced. With 1.5×10^{-4} M $[Hg^{2+}]$ (at 2.5×10^{-3} M $[Fe(CN)_6^{4-}]$), the rate falls rapidly, probably due to the formation of the 1:1 complex or higher complexes between $[Fe(CN)_6]^{4-}$ and $HgCl_2$. This complexation removes the catalytic species, viz, Hg^{2+} and $HgCl^+$ from the reaction mixture and causes a sharp decline in the rate. A similar observation has been reported by

us [23–25]. This supposition is in line with an earlier work of Beck [36] who demonstrated the formation of 1:1 complexes in reactions of $Hg(CN)_2$ with some inert cyano-complexes such as $[Fe(CN)_6^{4-} \cdot Hg(CN)_2]$, $[Fe(CN)_6^{3-} \cdot Hg(CN)_2]$, and $[Fe(CN)_6^{2-} \cdot Hg(CN)_2]$. However, another compound $Hg_2Fe(CN)_2$ is formed when $Hg(NO_3)_2$ is added to $[Fe(CN)_6]^{4-}$ solution [39]. According to Beck [36], the first complex may be represented as



Thus, the catalytic role of Hg^{2+} in the suggested mechanistic scheme appears to be justified. After an initial association between $[Fe(CN)_6]^{4-}$ and Hg^{2+}, the ion pair rapidly isomerizes to the following form:



The isomerized complex then decomposes in a slow step leading to the formation of $[Fe(CN)_5OH_2]^{3-}$ and $HgCN^+$. In the presence of INH, the water molecules of aquapentacyanoferrate(II) are quickly replaced leading to the formation of product $[Fe(CN)_5(INH)]^{3-}$. The repetitive spectral scans of the Hg(II)-catalyzed ligand substitution reaction between $[Fe(CN)_6]^{4-}$ and INH are shown in Fig. 7, which clearly reveals that a peak at 435 nm grows with time, which is attributed to the formation of a golden yellow complex $[Fe(CN)_5(INH)]^{3-}$ during the course of the reaction.

CONCLUSION

Based on our study, it is concluded that the antitubercular complex $[Fe(CN)_5(INH)]^{3-}$ is formed as a final reaction product during the Hg^{2+}-catalyzed ligand exchange reaction of INH via an interchange dissociative mechanism, which replaces one of the six coordinated cyanides in $[Fe(CN)_6]^{4-}$. The rate constants and other equilibrium constants involved in the mechanism have been evaluated. The computed activation parameters of the reaction are also provided in support of the proposed mechanism. The negative entropy of activation ($\Delta S^{\neq}$) for the reaction at hand is in contrast with the positive value found for other related $[Fe(CN)_5L]^{(3-n)-}$ complexes [34]. The decrease

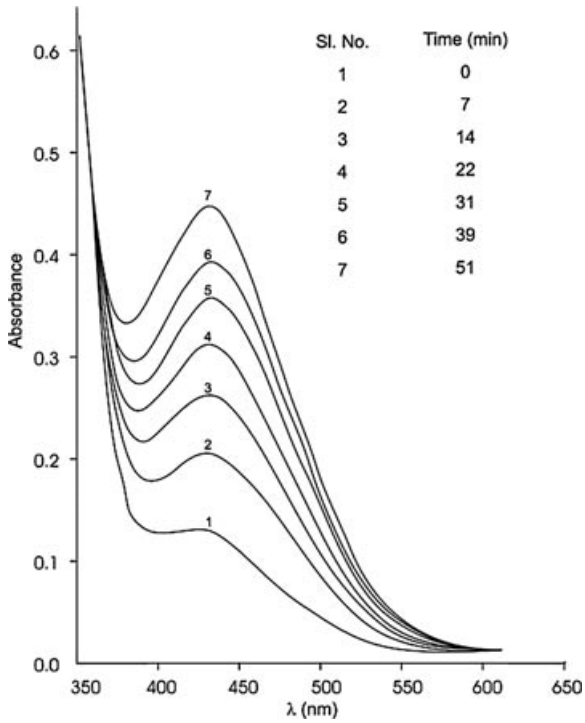


Figure 7 A repetitive spectral scan of the reaction mixture during a typical kinetic run at temperature = 25 ± 0.1°C, 2.5×10^{-3} M $[Fe(CN)_6^{4-}]$, 2.5×10^{-4} M [INH], $[Hg^{2+}] = 2.5 \times 10^{-6}$ M, pH 3.5 ± 0.02, $I = 0.05$ M (KNO_3), and temperature = 30.0 ± 0.1°C.

in the value of entropy of activation points toward a shift from a pure D-mechanism to an I_d-mechanism in $[Fe(CN)_6]^{4-}$. The overall mechanistic sequence described here is consistent with the kinetic studies of antitubercular complex formation.

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BIBLIOGRAPHY

1. Baraldo, L. M.; Forlano, P. F.; Parise, A. R.; Slep, L. D.; Olabe, J. A. Coord Chem Rev 2001, 219, 881.
2. Tobe, M. L.; Bergess, J. Inorganic Reaction Mechanism, Addition-Wesley-Longman: Essex, UK, 1999; Ch. 4.7.5.2 and 7.5.4.2.
3. Luiz, J. B.; Leigh, G. J.; Nunes, F. S. Polyhedron 2002, 21, 2137, and references therein.
4. Luiz, J. B.; de Andrade, F. M.; de Sa, E. L.; Friedermann, G. R.; Mangrich, A. S.; Barclay, J. E.; Evas, D. J.; Hasegawa, T.; Nunes, F. S. J Braz Chem Soc 2002, 15, 10, and references therein.
5. Toma, H. E.; Murakami, N. Y. Inorg Chem 1982, 21, 3573.
6. Burger, N.; Hankonyi, V. Anal Lett 1992, 25, 1355.

7. Stochel, G.; Chatlas, J.; Martinez, P.; van Eldik, R. Inorg Chem 1992, 31, 5480.
8. Stochel, G.; van Eldik, R. Inorg Chim Acta 1991, 190, 55.
9. Shifrin, N. S.; Beck, B. D.; Gauthier, T. D.; Chapnick, S. D.; Goodman, G. Regul Toxicol Pharmaco 1996, 23, 106.
10. Martell, A. E.; Smith, R. M. Critical Stability Constant; Inorganic Complexes; Plenum Press: New York, 1974.
11. Johnson, C. A.; Leinz, R. W.; Grimes, D. J.; Rye, R. O. Environ Sci Technol 2002, 36, 840, and references therein.
12. Bernstein, J.; Lott, W. A.; Steinberg, B. S.; Yale, H. L. Am Rev Tuberc 1952, 65, 357.
13. Zhang, H.; Wu, L.; Li, Q.; Du, X. Anal Chim Acta 2008, 628, 67.
14. Middlebrook, G. Am Rev Tuberc 1952, 65, 765.
15. Blanchard, J. S. Annu Rev Biochem 1996, 65, 215.
16. Basso, L. A.; Blanchard, J. S. Adv Exp Med Biol 1998, 456, 115.
17. Somoskovi, A.; Parsons, L. M.; Salfinger, M. Respir Res 2002, 2, 164.
18. Johnsson, K.; King, D. S.; Schultz, P. G. J Am Chem Soc 1995, 117, 5009.
19. Basso, L. A.; Zheng, R.; Blanchard, J. S. J Am Chem Soc 1996, 118, 11301.
20. Zabinski, R. F.; Blanchard, J. S. J Am Chem Soc 1997, 119, 2331.
21. Rozwarsky, D. A.; Grant, G. A.; Barton, D. H. R.; Jacobs. W. R.; Sacchettini, J. C. Science 1998, 279, 98.
22. Heym, B.; Alzari, P. M.; Honore, N.; Cole, S. T. Mol Microbiol 1995, 15, 235.
23. Naik, R. M.; Tiwari, R. K.; Singh, P. K.; Yadav, S. B. S. Int J Chem Kinet 2007, 39, 447, and references therein.
24. Prasad, S. Transition Met Chem 2003, 28, 1.
25. Naik, R. M.; Tiwari, R. K.; Singh, P. K.; Tiwari, A. K.; Prasad, S. Transition Met Chem 2005, 30, 968, and references therein.
26. Naik, R. M.; Singh R.; Asthana, A. Int J Chem Kinet 2011, 43, 21.
27. Naik, R. M.; Tiwari, R. K.; Singh, P. K.; Yadav, S. B. S.; Asthana, A. Transition Met Chem 2008, 33, 615.
28. Naik, R. M.; Singh, P. K.; Rastogi, R.; Singh, R.; Agarwal, A. Ann Chim 2007, 97, 1169.
29. Khun, D. D.; Young, T. C. Chemosphere 2005, 60, 1222.
30. Bansal, V. K.; Kumar, R.; Prasad, R.; Prasad, S.; Niraj, J. Mol Catal A 2008, 284, 69.
31. Weast, R. C. CRC Handbook of Chemistry and Physics, 49th ed.; The Chemical Rubber Company: Cleveland, OH, 1996; D-79.
32. Sousa, J. R.; Diogenes, I. C. N.; Carralho, I. M. M.; Temperini, M. L.; Tanaka, A. A.; Moreira, I. S. Dalton Trans 2003, 11, 2231.
33. Oliveira, J. S.; Sousa, E. H. S.; Basso, L. A.; Palaci, M.; Dietze, R.; Santos, D. S.; Moreira, I. S. Chem Commun 2004, 312.
34. Eaton, W. A.; George, P.; Hanaria, G. I. J. Phys Chem 1967, 71, 2016.
35. Mukherji, R.; Dhor, B. B.; Banerji, R.; Mukhopadhyay, S. J Coord Chem 2006, 59, 1157.
36. Beck, M. T. In: Proc. XXICCC, Calcutta, India in Coordination Chemistry-20; Pergamon Press: Oxford, UK, 1979; p. 31.
37. Belevant, S. V. I.; Reschevitskii, B. I. Koord Khim 1979, 5, 27.
38. Christensen, J. J.; Izatt, R. M.; Eatough, D. Inorg Chem 1965, 4, 1278.
39. Bellomo, A. Talanta 1975, 22, 197.