

## Mechanistic investigation on the oxidation of kinetin by Ag(III) periodate complex in aqueous alkaline media: A kinetic approach

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**Abstract.** The oxidation of amino acid, kinetin (KNT) by diperiodatoargentate(III) (DPA) in alkaline medium at a constant ionic strength of  $0.5 \text{ mol dm}^{-3}$  was studied spectrophotometrically. The reaction between KNT and DPA in alkaline medium exhibits 1 : 3 stoichiometry (KNT : DPA). Intervention of free radicals was observed in the reaction. Based on the observed orders and experimental evidences, a mechanism involving the monoperiodatoargentate(III) (MPA) as the reactive oxidant species has been proposed. The products, furon-2-methanol and para-nitro-purine were identified by spot test and characterized by spectral studies. The rate constants and associated activation parameters for the proposed mechanism as well as the thermodynamic quantities for different equilibrium steps are reported and discussed.

**Keywords.** Kinetics of oxidation; mechanism; kinetin; diperiodatoargentate(III); thermodynamic parameters.

### 1. Introduction

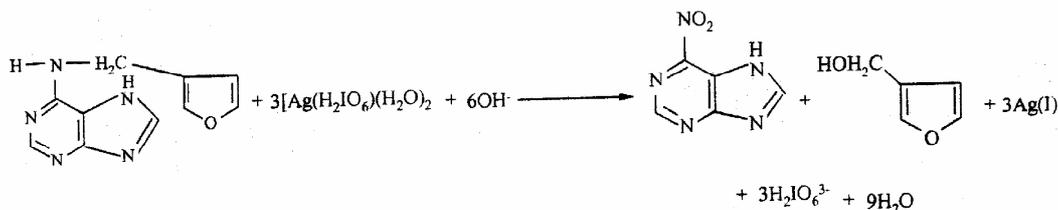
Kinetin is a kind of cytokinin, a class of plant hormone that promotes cell division. Kinetin exists naturally in the DNA of almost all organisms tested so far, including human cells, and various plants. The mechanism of production of KNT in DNA is thought to be via the production of furfural – an oxidative damage product of deoxyribose sugar in DNA – and its quenching by the adenine base converting it into N6-furfuryladenine, kinetin. Kinetin has been thoroughly tested for its powerful anti-aging effects in human skin cells and other systems. At present, kinetin is one of the widely used components in numerous skin care cosmetics and cosmeceuticals, such as valeant products kinerase.<sup>1</sup> There are some reports published on other biological effects of kinetin in human beings, for example, its effects as anti-platelet aggregation factor reducing thrombosis formation.

Diperiodatoargentate(III) (DPA) is a powerful oxidizing agent in alkaline medium with the reduction potential<sup>2</sup> 1.74 V. It is widely used as a volumetric reagent for the determination of various organic and inorganic species<sup>3,4</sup> Jayaprakash Rao

*et al*<sup>5</sup> have used DPA as an oxidizing agent for the kinetics of oxidation of various substrates. They normally found that order with respect to both oxidant and substrate was unity and  $[\text{OH}^-]$  was found to enhance the rate of reaction. It was also observed that they did not arrive at the possible active species of DPA in alkali and on the other hand, they proposed mechanism by generalizing the DPA as  $[\text{Ag}(\text{HL})\text{L}]^{(x+1)}$ . However Anil Kumar *et al*<sup>6,7</sup>, put an effort to give an evidence for the reactive form of DPA in the large scale of alkaline pH. In the present investigation, we have obtained the evidence for the reactive species for the DPA in alkaline medium. Since multiple equilibria between different Ag(III) species are involved, it would be interesting to know which of the species is the active oxidant.

In the later period of 20th century the kinetics of oxidation of various organic and inorganic substrate have been studied by Ag(III) species which may be due to its strong versatile nature of two electron oxidant. Among the various species of Ag(III),  $\text{Ag}(\text{OH})_4^-$ , diperiodatoargentate(III) and ethylenebis(biguanide) silver(III) (EBS) are of maximum attention to the researcher due to their relative stability.<sup>8</sup> The stability of  $\text{Ag}(\text{OH})_4^-$  is very sensitive towards traces of dissolved oxygen and other impurities in the reaction medium where upon it had not

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Scheme 1.

drawn much attention. However, the other forms of Ag(III)<sup>9,10</sup> are considerably stable; the DPA is used in highly alkaline medium and EBS is used in highly acidic medium.

In view of pharmaceutical and biological importance of KTN and lack of literature on the oxidation of KTN, it would be of interest to carry out a detailed investigation of the oxidation of KNT by DPA in aqueous alkaline medium (scheme 1). The thermodynamic quantities of various steps shown in scheme 1 have been computed to arrive at a suitable mechanism.

## 2. Experimental

All chemicals used were of reagent grade and millipore water was used throughout the work. A solution of KNT was prepared by dissolving an appropriate amount of recrystallised sample in millipore water. The purity of KNT was checked by comparing its melting point, 268°C with literature data (lit. m.p. 270°C). The required concentration of KNT was obtained from its stock solution. A stock solution of IO<sub>4</sub><sup>-</sup> was prepared by dissolving a known weight of KIO<sub>4</sub> (S.D. Fine) in hot water. The stock solution was used after keeping it for 24 h to complete the equilibrium. Its concentration was ascertained iodometrically<sup>11</sup> at neutral pH maintained by phosphate buffer. KNO<sub>3</sub> and KOH (BDH) were used to maintain ionic strength and alkalinity of the reaction respectively. An aqueous solution of AgNO<sub>3</sub> was used to study the product effect, Ag(I). The pH of the medium in the solution was measured by ELICO (LI613) pH meter.

### 2.1 Preparation of DPA

DPA was prepared by oxidizing Ag(I) in presence of KIO<sub>4</sub> as described elsewhere,<sup>12</sup> the mixture of 28 g of KOH and 23 g of KIO<sub>4</sub> in 100 cm<sup>3</sup> of water along with 8.5 g AgNO<sub>3</sub> was heated just to boiling and 20 g K<sub>2</sub>S<sub>2</sub>O<sub>8</sub> was added in several lots with stirring

and then allowed to cool. It was filtered through a medium porosity fritted glass filter and 40 g of NaOH was added slowly to the filtrate, where a voluminous orange precipitate agglomerated. The precipitate was filtered as above and washed three to four times with cold water to remove remaining K<sub>2</sub>S<sub>2</sub>O<sub>8</sub>. The pure crystals were dissolved in 50 cm<sup>3</sup> water and warmed to 80°C with constant stirring there by some solid was dissolved to give a red solution. The resulting solution was filtered when it was hot and on cooling at room temperature, the orange crystals separated out and were recrystallised from water, dried and stored in desiccator.

The complex was characterized from its UV-spectrum, which exhibited three peaks at 216, 255, and 362 nm. These spectral features were identical to those reported earlier<sup>12</sup> for DPA. The magnetic moment study revealed that the complex is diamagnetic. The compound prepared was analysed<sup>13</sup> for silver and periodate by acidifying a solution of the material with HCl, recovering and weighing the AgCl for Ag and titrating the iodine liberated when excess of KI was used for the required [DPA] solution in the reaction mixture.

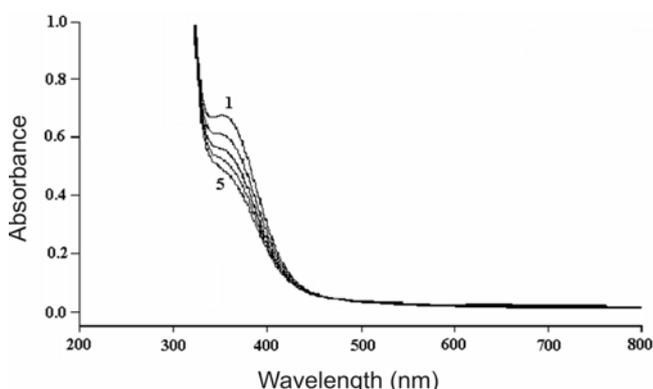
### 2.2 Kinetic studies

Since the initial reaction was too fast to be monitored by usual methods, kinetic measurements were performed on a Varian CARY Bio-50-UV-Vis spectrophotometer connected to a rapid kinetic accessory (HI-TECH SFA-12). The kinetics was followed under pseudo-first order condition where [KNT] > [DPA] at 25 ± 0.1°C unless specified. The reaction was initiated by mixing the DPA to KNT solution, which also contained required concentration of KNO<sub>3</sub>, KOH and KIO<sub>4</sub>. The progress of reaction was followed spectrophotometrically at 360 nm by monitoring the decrease in absorbance due to DPA with molar absorptivity index 'ε' to be 13900 ± 100 dm<sup>3</sup> mol<sup>-1</sup> cm<sup>-1</sup>. It was verified that there is a negligible interference from other

species present in the reaction mixture at this wavelength.

The reaction was followed to more than 85% completion of the reaction. Plots of log (absorbance) versus time lead to the first order rate constant ( $k_{\text{obs}}$ ). The plots were linear up to 85% completion of reaction and rate constants were reproducible within  $\pm 5\%$ . During the kinetics, a constant concentration ( $1.0 \times 10^{-5} \text{ mol dm}^{-3}$ ) of  $\text{KIO}_4$  was used throughout the study unless otherwise stated. Thus, possibility of oxidation of KNT by periodate was tested and found that there was no significant interference due to  $\text{KIO}_4$  under experimental condition. The total concentration of  $\text{OH}^-$  was calculated by considering the amount present in the DPA solution and additionally added. Kinetic runs were also carried out in  $\text{N}_2$  atmosphere in order to understand the effect of dissolved oxygen on the rate of reaction. No significant difference in the results was obtained under a  $\text{N}_2$  atmosphere and in the presence of air. In view of the ubiquitous contamination of carbonate in the basic medium, the effect of carbonate was also studied. Added carbonate had no effect on the reaction rates. The spectral changes during the reaction are shown in figure 1. It is evident from the figure 1 that the concentration of DPA decreases at 360 nm.

In view of the modest concentration of alkali used in the reaction medium, attention was also directed to the effect of the reaction vessel surface on the kinetics. Use of polythene acrylic wares and quartz or polyacrylate cells gave the same results, indicating that the surface did not have any significant effect on the reaction rates. Regression analysis of experimental data to obtain regression coefficient



**Figure 1.** Spectroscopic changes occurring in the oxidation of KNT by DPA at 298 K,  $[\text{DPA}] = 5.0 \times 10^{-5}$ ,  $[\text{KNT}] = 5.0 \times 10^{-4}$ ,  $[\text{OH}^-] = 0.50$ ; and  $I = 0.50 \text{ mol dm}^{-3}$  with scanning time interval of (1) 1.0, (2) 2.0, (3) 3.0, (4) 4.0, (5) 5.0 min.

$r$  and the standard deviation  $S_r$  of points from the regression line, was performed with the Microsoft Office Excel 2003 programme.

### 3. Results

#### 3.1 Stoichiometry and product analysis

Different sets of reaction mixtures containing varying ratios of DPA to kinetin in presence of constant amount of  $\text{OH}^-$ ,  $\text{KNO}_3$  were kept for 4 h in closed vessel under nitrogen atmosphere. The remaining concentration of DPA was estimated by spectrophotometrically at 360 nm. The results indicated a 1 : 3 stoichiometry as given in scheme 2.

The main oxidation products were identified as para-nitro-purine and furon-2-methanol, and were characterized by its melting point, IR and GC-MS respectively. The nature of furon-2-methanol was confirmed by its IR spectrum, which showed a  $-\text{OH}$  stretch at  $3386 \text{ cm}^{-1}$  indicating the presence of alcohol and para-nitro-pteridine was also confirmed by the presence of  $\text{NO}_2$  group, stretching frequency at  $1332 \text{ cm}^{-1}$ . Further, the products were subjected to GC-mass spectral analysis. GC-MS data was obtained on a QP-2010S Shimadzu gas chromatograph mass spectrometer. The mass spectral data showed a base peak at 98 m/z thus, confirming the presence of furon-2-methanol (figure 2) and another product, was also confirmed by mass spectral data which showed a peak at 182 m/z, hence confirming the product para-nitro-pteridine (figure 3). All other peaks observed in GC-MS can be interpreted in accordance with the observed structure of para-nitro-pteridine and furon-2-methanol.

The by-products were identified as ammonia by Nessler's reagent,<sup>14</sup> the  $\text{CO}_2$  was qualitatively detected by bubbling nitrogen gas through the acidified reaction mixture and passing the liberated gas through tube containing lime water. The formation of free  $\text{Ag}^+$  in solution was detected by adding KCl solution to the reaction mixture, which produced white turbidity due to formation of  $\text{AgCl}$ . It was observed that para-nitro-pteridine and furon-2-methanol did not undergo further oxidation under the present kinetic condition.

#### 3.2 Reaction orders

The reaction orders were determined from the slope of  $\log k_{\text{obs}}$  vs  $\log$  (concentration) plots by varying the

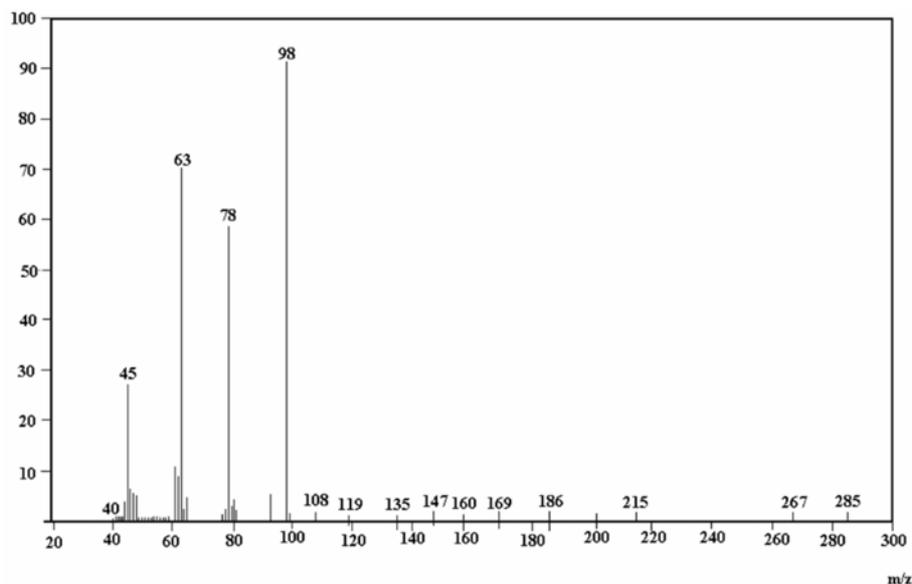


Figure 2. Mass spectrum of reaction product, furon-2-methanol at 98 amu.

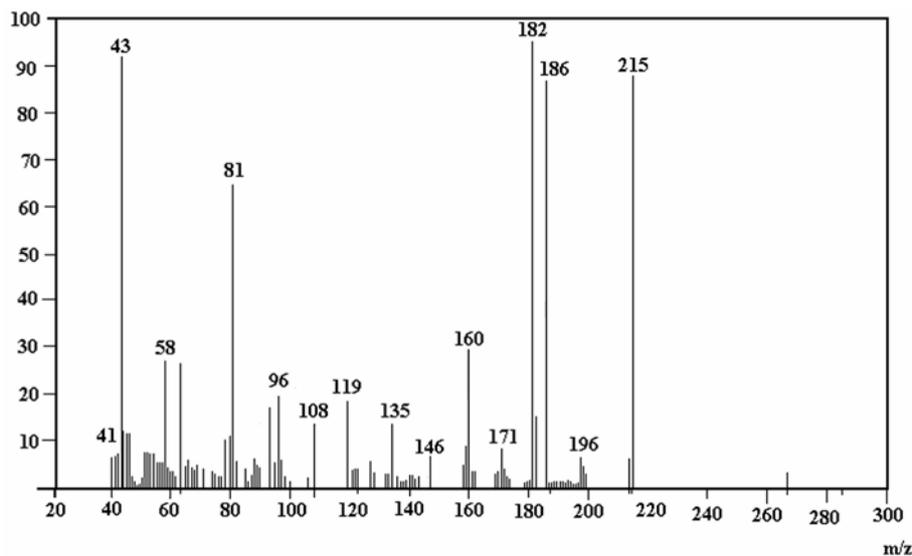


Figure 3. Mass spectrum of reaction product, para-nitro-pteridine at 182 amu.

concentration of kinetin, alkali in turn while keeping all other concentrations and conditions constant.

### 3.3 Effect of [diperiodatoargentate (III)]

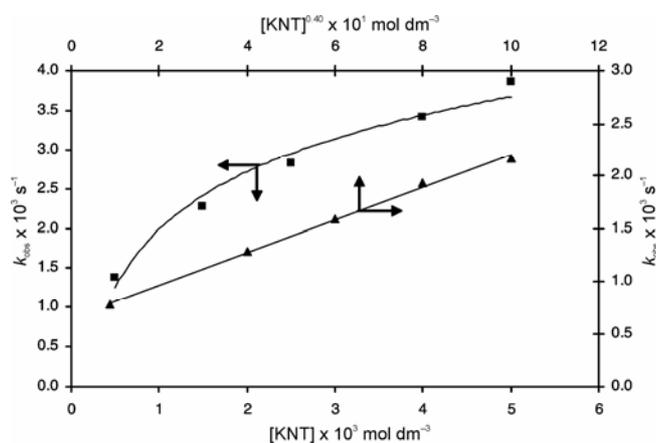
The oxidant DPA concentration was varied in the range of  $1.0 \times 10^{-5}$  to  $1.0 \times 10^{-4}$  mol dm<sup>-3</sup> and fairly constant  $k_{\text{obs}}$  value indicate that order with respect to [DPA] was unity (table 1). This was also confirmed by linearity of the plots of log (absorbance) vs time to 85% completion of reaction.

### 3.4 Effect of [kinetin]

The effect of KNT on the rate of reaction was studied at constant concentration of alkali, DPA and periodate at constant ionic strength of 0.50 mol dm<sup>-3</sup>. The substrate KNT was varied in the range of  $1.0 \times 10^{-4}$  to  $1.0 \times 10^{-3}$  mol dm<sup>-3</sup>. The  $k_{\text{obs}}$  values increased with increase in concentration of KNT. The apparent order with respect to [KNT] was found to be less than unity (table 1) ( $r \geq 0.998$ ,  $S \leq 0.02$ ). This was also confirmed by the plots of  $k_{\text{obs}}$  vs [KNT]<sup>0.4</sup> which is linear rather than the direct plot of  $k_{\text{obs}}$  vs [KNT] (figure 4).

**Table 1.** Effect of variation of [DPA], [KNT], [OH<sup>-</sup>] and [IO<sub>4</sub><sup>-</sup>] on the oxidation of kinetin by diperiodatoargentate(III) in aqueous alkaline medium at 298 K and I = 0.50 mol dm<sup>-3</sup>.

10 <sup>5</sup> [DPA] (mol dm <sup>-3</sup> )	10 <sup>4</sup> [KNT] (mol dm <sup>-3</sup> )	[OH <sup>-</sup> ] (mol dm <sup>-3</sup> )	10 <sup>5</sup> [IO <sub>4</sub> <sup>-</sup> ] (mol dm <sup>-3</sup> )	10 <sup>3</sup> k <sub>obs</sub>	10 <sup>3</sup> k <sub>cal</sub>
1.0	5.0	0.5	1.0	2.2	2.0
3.0	5.0	0.5	1.0	2.1	2.0
5.0	5.0	0.5	1.0	2.0	2.0
8.0	5.0	0.5	1.0	2.3	2.0
10.0	5.0	0.5	1.0	2.1	2.0
5.0	1.0	0.5	1.0	0.9	0.87
5.0	3.0	0.5	1.0	1.6	1.6
5.0	5.0	0.5	1.0	2.0	1.9
5.0	8.0	0.5	1.0	2.3	2.2
5.0	10.0	0.5	1.0	2.5	2.4
5.0	5.0	0.05	1.0	0.69	0.68
5.0	5.0	0.1	1.0	1.1	1.0
5.0	5.0	0.2	1.0	1.6	1.4
5.0	5.0	0.3	1.0	1.8	1.7
5.0	5.0	0.5	1.0	2.0	1.9
5.0	5.0	0.5	0.5	2.4	2.4
5.0	5.0	0.5	0.8	2.2	2.2
5.0	5.0	0.5	1.0	2.0	1.9
5.0	5.0	0.5	3.0	1.4	1.4
5.0	5.0	0.5	5.0	1.2	1.1

**Figure 4.** Plots of k<sub>obs</sub> vs [KNT]<sup>0.40</sup> and k<sub>obs</sub> vs [KNT] (conditions as in table 1).

### 3.5 Effect of [alkali]

The effect of increase in concentration of alkali on the reaction was studied at constant concentration of KNT, DPA and periodate at constant ionic strength of 0.50 mol dm<sup>-3</sup> at 25°C. The rate constants increased with increase in alkali concentrations (table 1), indicating positive fractional order dependence of rate on alkali concentration ( $r \geq 0.996$ ,  $S \leq 0.02$ ).

### 3.6 Effect of [periodate]

The effect of increasing concentration of periodate was studied by varying the periodate concentration from  $1.0 \times 10^{-5}$  to  $1.0 \times 10^{-4}$  mol dm<sup>-3</sup> keeping all other reactants concentration constant. It was found that added periodate had retarding effect on the rate of reaction.

### 3.7 Effect of ionic strength (I) and dielectric constant of medium (D)

The addition of KNO<sub>3</sub> at constant [DPA], [KNT], [OH<sup>-</sup>] and [IO<sub>4</sub><sup>-</sup>] was found that increasing ionic strength of the reaction medium did not effect the rate of reaction. Varying the *t*-butyl alcohol and water percentage varied dielectric constant of the medium 'D'. The *D* values were calculated from the equation  $D = D_w V_w + D_B V_B$ , where *D<sub>w</sub>* and *D<sub>B</sub>* are dielectric constants of pure water and *t*-butyl alcohol respectively and *V<sub>w</sub>* and *V<sub>B</sub>* are the volume fractions of components of water and *t*-butyl alcohol respectively in the total mixture. The decrease in dielectric constant of the reaction medium had no effect on the rate of reaction.

### 3.8 Effect of initially added products

The externally added product Ag(I), para-nitro-pteridine and furon-2-methanol did not have any significant effect on the rate of reaction.

### 3.9 Polymerization study

The intervention of free radicals in the reaction was examined as follows: The reaction mixture, to which a known quantity of acrylonitrile monomer initially added, was kept for 2 h in an inert atmosphere. On

**Table 2.** Activation parameters and thermodynamic quantities for the oxidation of KNT by diperiodatoargentate(III) in aqueous alkaline medium with respect to the slow step of scheme 2.

(a) Effect of temperature	
Temperature (K)	$10^3 k$ (s <sup>-1</sup> )
293	2.2
298	2.8
303	3.9
308	4.7

(b) Activation parameters	
Parameter	Value
$E_a$ (kJ mol <sup>-1</sup> )	$39.4 \pm 2$
$\Delta H^\ddagger$ (kJ mol <sup>-1</sup> )	$37 \pm 21$
$\Delta S^\ddagger$ (JK <sup>-1</sup> mol <sup>-1</sup> )	$-169 \pm 10$
$\Delta G^\ddagger$ (kJ mol <sup>-1</sup> )	$87 \pm 4$
$\log A$	$4.3 \pm 0.2$

(c) Effect of temperature on  $K_1$ ,  $K_2$  and  $K_3$  for the oxidation of KNT by diperiodatoargentate(III) in aqueous alkaline medium.

Temperature (K)	$K_1$ (dm <sup>3</sup> mol <sup>-1</sup> )	$K_2 \times 10^5$ (mol dm <sup>-3</sup> )	$K_3 \times 10^{-4}$ (dm <sup>3</sup> mol <sup>-1</sup> )
293	0.3	1.0	2.6
298	0.4	1.4	2.3
303	0.5	1.6	1.9
308	0.6	1.8	1.6

(d) Thermodynamic quantities using  $K_1$ ,  $K_2$  and  $K_3$

Thermodynamic quantities	Values from $K_1$	Values from $K_2$	Values from $K_3$
$\Delta H$ (kJ mol <sup>-1</sup> )	$35.3 \pm 4.0$	$28.5 \pm 0.7$	$-23.8 \pm 0.8$
$\Delta S$ (JK <sup>-1</sup> mol <sup>-1</sup> )	$111.1 \pm 23$	$117.1 \pm 13$	$118.3 \pm 22$
$\Delta G$ (kJ mol <sup>-1</sup> )	$2.0 \pm 0.12$	$-6.6 \pm 0.3$	$-59.3 \pm 3.1$

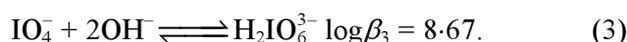
diluting the reaction mixture with methanol, a white precipitate was formed, which indicated the intervention of free radicals in the reaction.<sup>15</sup>

### 3.10 Effect of temperature

The kinetics was studied at four different temperatures (15, 20, 25 and 30°C) under varying concentrations of KNT, alkali and periodate keeping all other conditions constant. The rate constants ( $k$ ), of the slow step of the reaction mechanism were obtained from the slopes and intercepts of the plots of  $1/k_{\text{obs}}$  vs  $1/[\text{KNT}]$ ,  $1/k_{\text{obs}}$  vs  $1/[\text{OH}^-]$  and  $1/k_{\text{obs}}$  vs  $[\text{H}_3\text{IO}_6^{2-}]$  at four different temperatures. The values are given in table 2.

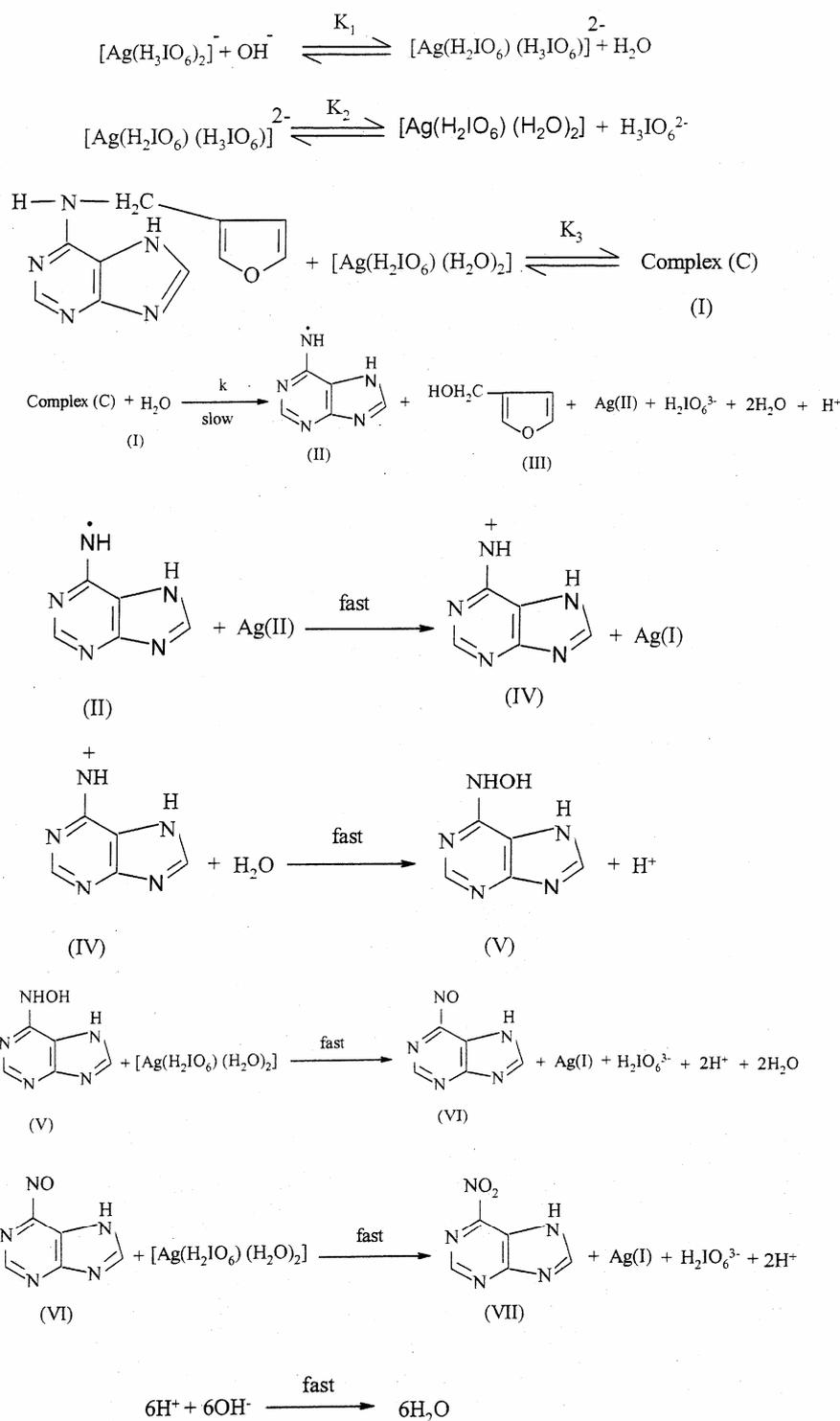
## 4. Discussion

The literature survey<sup>12</sup> reveals that the water soluble diperiodatoargentate(III) (DPA) has a formula  $[\text{Ag}(\text{IO}_6)_2]^{7-}$  with  $dsp^2$  configuration of square planar structure, similar to diperiodatocopper(III) complex with two bidentate ligands, periodate to form a planar molecule. In the alkaline medium, the dissociative equilibria (1–3) of the  $\text{IO}_4^-$  were detected and the corresponding equilibrium constants were determined at 298.2 K by Aveston.<sup>16</sup>



The distribution of all species of periodate in aqueous alkaline solution can be calculated from equilibria (1–3). In the  $[\text{OH}^-]$  range used in this work the amount of dimer ( $\text{H}_2\text{I}_2\text{O}_{10}^{4-}$ ) and  $\text{IO}_4^-$  species of periodate can be neglected. The main species of periodate are  $\text{H}_2\text{IO}_6^{3-}$  and  $\text{H}_3\text{IO}_6^{2-}$ , which are consistent with the result calculated from Crouthamel's data.<sup>17</sup> Hence DPA could be as  $[\text{Ag}(\text{H}_3\text{IO}_6)_2]^-$  or  $[\text{Ag}(\text{H}_2\text{IO}_6)_2]^{3-}$  in alkaline medium. Therefore, under the present condition, diperiodatoargentate(III), may be depicted as  $[\text{Ag}(\text{H}_3\text{IO}_6)_2]^-$ . The similar speciation of periodate in alkali was proposed<sup>18</sup> for diperiodatonickelate(IV).

The reaction between diperiodatoargentate(III) complex and kintin in alkaline medium has the stoichiometry 1 : 3 (KNT : DPA) with a first order



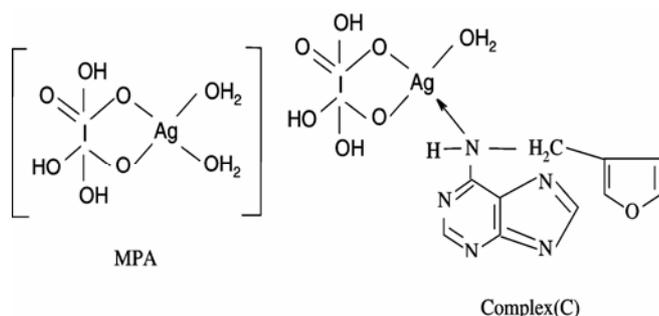
**Scheme 2.** Detailed scheme for the oxidation of KNT by alkaline DPA.

dependence on [DPA] and an apparent order of less than unit order in [substrate], [alkali] and negative fractional order in [periodate]. No effect of added products was observed. Based on the experimental results, a mechanism is proposed for which all the

observed orders in each constituent such as [oxidant],  $[\text{OH}^-]$ , and  $[\text{IO}_4^-]$  may be well accommodated (scheme 2).

In view of the observed experimental results, monoperiodatoargentate(III) (MPA) is considered to

be the active species. The fractional order dependence of  $k_{\text{obs}}$  on  $[\text{OH}^-]$  suggests that  $\text{OH}^-$  takes part in the pre-equilibrium step 1 with DPA to give a deprotonated diperiodatoargentate(III). The plot of  $1/k_{\text{obs}}$  vs  $[\text{IO}_4^-]$  is linear with positive intercept indicating a dissociative equilibrium in which the DPA loses a periodate ligand from its coordination sphere, forming a reactive monoperiodatoargentate(III) complex (MPA) in the second step, which is evidenced by decrease in the rate with increase in  $[\text{IO}_4^-]$ . It may be expected that lower Ag(III) periodate species such as MPA will be more important in the reaction than DPA. The inverse fractional order in  $\text{H}_3\text{IO}_6^{2-}$  concentration might also due to this reason. The fractional order with respect to KNT presumably results from the complex formation between MPA and KNT prior to the slow step. Indeed it is to be noted that a plot of  $1/k_{\text{obs}}$  vs  $1/[\text{KNT}]$  was linear and shows an intercept in agreement with the complex formation. The KNT-DPA complex (I) is formed by chelation of DPA molecule at amine group of KTN. As oxygen is more electronegative compared to nitrogen, nitrogen donates a lone pair of electron to monoperiodatoargentate(III). This complex (I) decomposes in a slow step to give 1H-imidazole pyridine-4-yl amine radical (II), furon-3-yl-methanol (III) and Ag(II) species. Imidazole pyridine 4-yl-amine radical (II) combines with Ag(II) species in a fast step to give 1H-imidazole pyridine 4-yl-amine cation (IV) and Ag(I) species. This amine cation (IV) reacts with water molecule in a fast step, to give 1H imidazole N-pyrimidine 4-yl-hydroxyl amine (V), which further reacts with another mole of MPA in a fast step to give 1H-imidazole 4-nitroso-pyrimidine (VI) and Ag(I) species. In further fast step the 1H-Imidazole-4-nitroso pyrimidine (VI) reacts with another molecule of MPA to give the product 1H-imidazole-nitro pyrimidine (VII) and Ag(I). All these results may be interpreted in the form of scheme 2. The probable structure of MPA and complex(C) (I) are given below.



Spectroscopic evidence for the complex formation between oxidant and substrate was obtained from UV-Visible spectra of  $[\text{DPA}]$  ( $5.0 \times 10^{-5} \text{ mol dm}^{-3}$ ),  $[\text{KNT}]$  ( $5.0 \times 10^{-4} \text{ mol dm}^{-3}$ ),  $[\text{OH}^-]$  ( $0.5 \text{ mol dm}^{-3}$ ) and mixture of DPA and kinetin. A bathochromic shift of about 5 nm from 234 to 239 nm in the spectra of DPA to mixture of substrate and DPA was observed. However, the Michaelis-Menten plot proved the complex formation between DPA and kinetin. Such type of complex between an oxidant and substrate has also been observed in other studies<sup>19</sup> scheme 2 leads to the rate law (4)

$$\text{Rate} = \frac{kK_1K_2K_3[\text{DPA}][\text{OH}^-][\text{KNT}]}{\left[ [\text{H}_3\text{IO}_6^{2-}] + K_1[\text{OH}^-][\text{H}_3\text{IO}_6^{2-}] + K_1K_2[\text{OH}^-] + K_1K_2K_3[\text{OH}^-][\text{KNT}] \right]}, \quad (4)$$

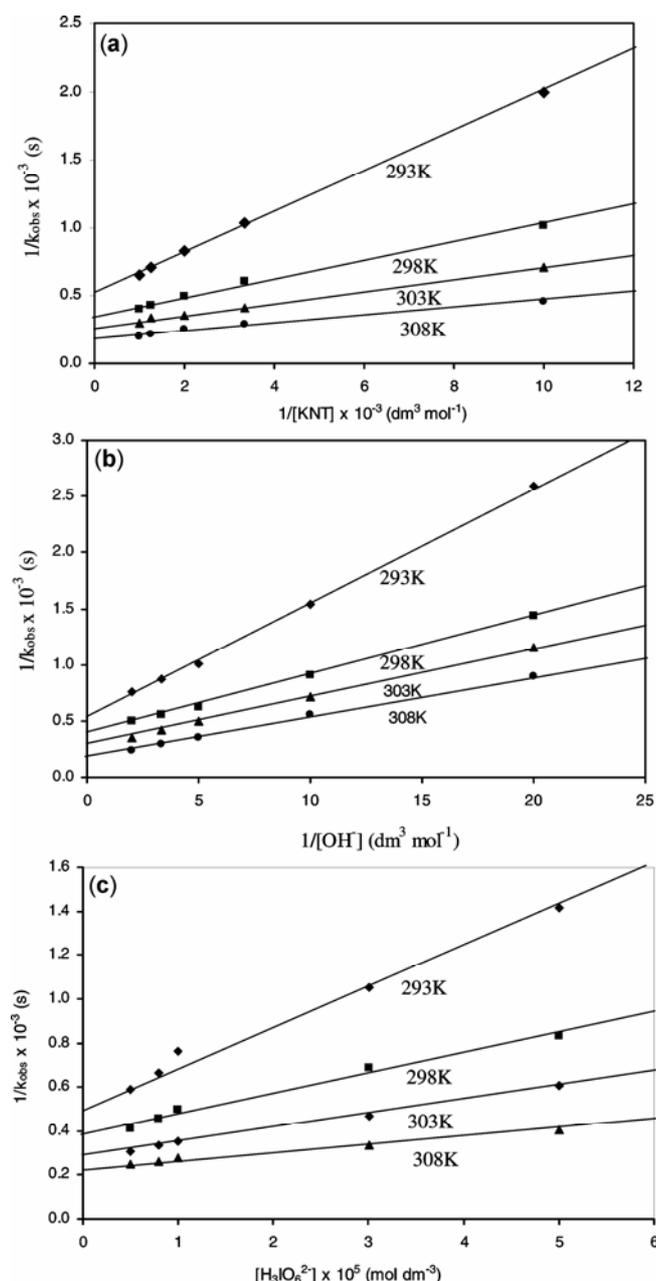
$$k_{\text{obs}} = \frac{\text{Rate}}{[\text{DPA}]} = \frac{kK_1K_2K_3[\text{OH}^-][\text{KNT}]}{\left[ [\text{H}_3\text{IO}_6^{2-}] + K_1[\text{OH}^-][\text{H}_3\text{IO}_6^{2-}] + K_1K_2[\text{OH}^-] + K_1K_2K_3[\text{OH}^-][\text{KNT}] \right]} \quad (5)$$

This explains all the observed kinetic orders of different species. The rate law (5) can be rearranged in the following form, which is suitable for verification.

$$\frac{1}{k_{\text{obs}}} = \frac{[\text{H}_3\text{IO}_6^{2-}]}{kK_1K_2K_3[\text{OH}^-][\text{KNT}]} + \frac{[\text{H}_3\text{IO}_6^{2-}]}{kK_2K_3[\text{KNT}]} + \frac{1}{kK_3[\text{KNT}]} + \frac{1}{k} \quad (6)$$

According to (6), other conditions being constant, plots of  $1/k_{\text{obs}}$  vs  $1/[\text{KNT}]$  ( $r \geq 0.9998$ ,  $S \leq 0.016$ ),  $1/k_{\text{obs}}$  vs  $1/[\text{OH}^-]$  ( $r \geq 0.996$ ,  $S \leq 0.019$ ) and  $1/k_{\text{obs}}$  vs  $[\text{H}_3\text{IO}_6^{2-}]$  ( $r \geq 0.9312$ ,  $S \leq 0.017$ ) should be linear and are found to be so (figures 5a-c). The slopes and intercepts of such plots leads to the value of  $K_1$ ,  $K_2$ ,  $K_3$  and  $k$  as  $0.40 \text{ dm}^3 \text{ mol}^{-1}$ ,  $1.4 \times 10^{-5} \text{ mol dm}^{-3}$ ,  $2.3 \times 10^4 \text{ dm}^3 \text{ mol}^{-1}$  and  $2.8 \times 10^{-3} \text{ s}^{-1}$  at 298 K and  $I = 0.5 \text{ mol dm}^{-3}$  respectively. The values of  $K_1$  and  $K_2$  are in good agreement with earlier literature.<sup>20</sup> These constants were used to calculate the rate constant and compared with the experimental  $k_{\text{obs}}$  values and found to be in reasonable agreement with each other which fortifies scheme 2.

The thermodynamic quantities for the first, second and third equilibrium steps of scheme 2 can be evaluated as follows:  $[KNT]$ ,  $[OH^-]$  and  $[IO_4^-]$  (table 1) were varied at four different temperatures. The plot of  $1/k_{obs}$  vs  $1/[KNT]$  ( $r \geq 0.9998$ ,  $S \leq 0.016$ )  $1/k_{obs}$  vs  $1/[OH^-]$  ( $r \geq 0.996$ ,  $S \leq 0.019$ ), and  $1/k_{obs}$  vs  $[H_3IO_6^{2-}]$  ( $r \geq 0.9912$ ,  $S \leq 0.017$ ) should be linear.



**Figure 5.** Verification of rate law (4) for the oxidation of KNT by DPA. (a)  $1/k_{obs}$  vs  $1/[KNT]$  at four different temperatures (condition as in table 1). (b)  $1/k_{obs}$  vs  $1/[OH^-]$  at four different temperatures (condition as in table 1). (c)  $1/k_{obs}$  vs  $[H_3IO_6^{2-}]$  at four different temperatures (condition as in table 1).

From the slopes and intercepts, the values of  $K_1$ ,  $K_2$  and  $K_3$  were calculated at four different temperatures and these values are given in table 2. The vant Hoff's plots were made for variation of  $K_1$ ,  $K_2$  and  $K_3$  with temperatures ( $\log K_1$  vs  $1/T$ ) ( $r \geq 0.9949$ ,  $S \leq 0.00898$ ) ( $\log K_2$  vs  $1/T$ ) ( $r \geq 0.9362$ ,  $S \leq 0.006$ ) and ( $\log K_3$  vs  $1/T$ ) ( $r \geq 0.9985$ ,  $S \leq 0.006$ ) and the values of enthalpy of reaction  $\Delta H$ , entropy of reaction  $\Delta S$  and free energy of reaction  $\Delta G$ , were calculated for the first, second and third equilibrium steps. These values are given in table 2. A comparison of  $\Delta H$  ( $35.3 \pm 0.8$   $kJ mol^{-1}$ ) from  $K_1$  with that of  $\Delta H^\ddagger$  ( $37.0 \pm 1.0$   $kJ mol^{-1}$ ), the rate limiting step, supporting the fact that the first step of scheme 2 is fairly fast since it involves a low activation energy.<sup>21</sup>

The negligible effect of ionic strength and dielectric constant might be due to involvement of neutral substrate in the reaction (scheme 2). The values  $\Delta H^\ddagger$  and  $\Delta S^\ddagger$  were both favourable for electron transfer processes. The favourable enthalpy was due to release of energy on solutions changes in the transition state. The negative value of  $\Delta S^\ddagger$  suggests that the intermediate complex is more ordered than the reactant.<sup>22</sup> The observed modest enthalpy of activation and a higher rate constant for the slow step indicates that the oxidation presumably occurs via an inner-sphere mechanism. This conclusion is supported by earlier observations.<sup>21</sup>

## 5. Conclusion

Among the various species of DPA in alkaline medium, MPA, i.e.  $[Ag(H_2IO_6)(H_2O)_2]$  is considered as active species for the title reaction. The results indicated that, the role of pH in the reaction medium is crucial. Rate constant of slow step and other equilibrium constants involved in the mechanism were evaluated and activation parameters with respect to slow step of reaction were computed. The overall mechanistic sequence described is consistent with product studies, mechanistic and kinetic studies.

## Appendix I

Derivation of rate equation:

According to scheme 2,

$$\text{Rate} = k[C] = \frac{kK_1K_2K_3[KNT][DPA][OH^-]}{[H_3IO_6]^{2-}} \quad (I)$$

$$\begin{aligned}
 [\text{DPA}]_T &= [\text{DPA}]_f + [\text{Ag}(\text{H}_3\text{IO}_6)(\text{H}_2\text{IO}_6)]^{2-} \\
 &\quad + [\text{Ag}(\text{H}_3\text{IO}_6)(\text{H}_2\text{O})_2] + \text{C} \\
 &= [\text{DPA}]_f + \left[ \frac{1 + K_1[\text{OH}^-] + \frac{K_1K_2[\text{OH}^-]}{[\text{H}_3\text{IO}_6^{2-}]} + \frac{K_1K_2K_3[\text{KNT}][\text{OH}^-]}{[\text{H}_3\text{IO}_6^{2-}]}}{[\text{H}_3\text{IO}_6^{2-}]} \right],
 \end{aligned}$$

where  $T$  and  $f$  refer to total and free concentrations

$$[\text{DPA}]_f = \frac{[\text{DPA}][\text{H}_3\text{IO}_6^{2-}]}{[\text{H}_3\text{IO}_6^{2-}] + K_1[\text{OH}^-][\text{H}_3\text{IO}_6^{2-}] + K_1K_2[\text{OH}^-] + K_1K_2K_3[\text{OH}^-][\text{KNT}]} \quad (\text{II})$$

$$\begin{aligned}
 [\text{OH}]_T &= [\text{OH}]_f + [\text{Ag}(\text{H}_3\text{IO}_6)(\text{H}_2\text{IO}_6)]^{2-} \\
 &\quad + [\text{Ag}(\text{H}_3\text{IO}_6)(\text{H}_2\text{O})_2] \\
 &= [\text{OH}^-]_f + K_1[\text{DPA}][\text{OH}^-] + \frac{K_1K_2[\text{DPA}][\text{OH}^-]}{[\text{H}_3\text{IO}_6^{2-}]}.
 \end{aligned}$$

In view of the low concentration of  $[\text{DPA}]$  and  $[\text{H}_3\text{IO}_6^{2-}]$  used:

$$[\text{OH}^-]_T = [\text{OH}^-]_f. \quad (\text{III})$$

Similarly,

$$\begin{aligned}
 [\text{KNT}]_T &= [\text{KNT}]_f + [\text{C}] \\
 &= [\text{KNT}]_f = \frac{K_1K_2K_3[\text{KNT}][\text{DPA}][\text{OH}^-]}{[\text{H}_3\text{IO}_6^{2-}]}.
 \end{aligned}$$

In view of the low concentration of  $[\text{DPA}]$ ,  $[\text{OH}^-]$ , and  $[\text{H}_3\text{IO}_6^{2-}]$  used:

$$[\text{KNT}]_T = [\text{KNT}]_f. \quad (\text{IV})$$

Substituting (II), (III), (IV) in (I) and omitting the subscripts, we get,

$$\text{Rate} = \frac{kK_1K_2K_3[\text{DPA}][\text{KNT}][\text{OH}^-]}{[\text{H}_3\text{IO}_6^{2-}] + K_1[\text{OH}^-][\text{H}_3\text{IO}_6^{2-}] + K_1K_2[\text{OH}^-] + K_1K_2K_3[\text{OH}^-][\text{KNT}]}.$$

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