Chapter III

Osmium(VIII) Catalyzed Oxidation of DL-Methionine by Hexacyanoferrate (III) in Aqueous Alkaline Medium
Methionine, is a sulfur containing essential amino acid which is not synthesized in the body and must be obtained from food. It contributes to supply mineral sulfur improving the tone and pliability of the skin, conditioning the hair and strengthens nails and protecting the cells from airborne pollutants. It contributes to other compounds including S-adenosyl-methionine (SAM), which participate in liable methyl groups and sulfur to over 100 biochemical reactions for normal brain function, and L-cysteine, which is a component of glutathione, an important antioxidant molecule in the body. Methionine is also a transporter of the antioxidant mineral selenium. Methionine occurs in two optical isomers, a dextro and a levo form.
The general importance of hexacyanoferrate(III), ([Fe(CN)₆]³⁻) is given in Chapter II (p.46).

Osmium(VIII) has long been used as a catalyst in redox reactions¹ and as an oxidizing agent². Its action in alkaline medium has been more widely studied and is better understood than its action in acid. However, in most of osmium(VIII) catalysis it involves in oxidation of substrate followed by regeneration with the interaction of oxidant. In the present study its role is entirely different.

Several studies have been reported on the oxidation of DL-methionine (DLM) by other oxidants such as KMnO₄³, Ce(IV)⁴, and Cr(III)⁵ and Pyridinium Fluorochromate⁶. Different workers have identified different products by different oxidants for DL-methionine. Thus, the study of DL-methionine becomes important because of its biological significance and selectivity towards the oxidants. In view of the lack of literature on the oxidation of DL-methionine by [Fe(CN)₆]³⁻ and in order to explore the mechanistic aspects of [Fe(CN)₆]³⁻ oxidation in alkaline medium, we have chosen DL-methionine as a substrate and it is an interesting aspect to note that the uncatalyzed reaction is very very slow. However, with a trace amount of osmium(VIII), reaction rate increases by nearly thousand folds and also earlier reports¹ reveal that the kinetics of osmium(VIII) catalysed oxidation of α-amino acids by [Fe(CN)₆]³⁻ under first order reaction condition, the plots of log (concentration) versus time were not linear due to strong catalytic influence of [Fe(CN)₆]⁴⁺. As the [Fe(CN)₆]⁴⁺ is a reduction product of [Fe(CN)₆]³⁻, its accelerating influence was encountered in every kinetic run which
showed curves with increasing slopes. In order to understand the oxidation of DL-methionine, the role osmium(VIII) as a catalyst and influence of the product, $[\text{Fe(CN)}_6]^{4-}$, the title work is undertaken for the investigation.

**EXPERIMENTAL**

**Materials and Methods**

All chemicals used were of reagent grade. Double distilled water was used throughout the work. A solution of $[\text{Fe(CN)}_6]^{3-}$ was prepared by dissolving $\text{K}_3\text{Fe(CN)}_6$ (BDH) in $\text{H}_2\text{O}$ and was standardized iodometrically$^7$. (Chaper II(p.48). DL-methionine is a colorless or white lustrous plates, or a white crystalline powder and has a slight characteristic odour, soluble in water, alkali and mineral acids, and partially soluble in alcohol but insoluble in ether. Stock solution of DL-methionine (s. d-FINE) was prepared by dissolving the appropriate amount of purified sample of DL-methionine sample in water. The purity of the sample was checked by TLC and its M.P. ($279^\circ\text{C}$). The stock solution of osmium(VIII) was obtained by dissolving osmium tetroxide (Johnson Matthey) in 0.50 mol dm$^{-3}$ sodium hydroxide solution and its concentration was ascertained$^8$ by standardizing it with standard ceric ammonium sulphate solution. $\text{NaOH}$ (Merk) and $\text{NaClO}_4$ (BDH) were used to provide the required alkalinity and to maintain the ionic strength, respectively in reaction solutions.
Kinetics

All kinetic measurements were performed under pseudo-first order conditions with [DL-methionine] in excess over [Fe(CN)₆]³⁻ at constant ionic strength of 0.5 mol dm⁻³. The reaction was initiated by mixing previously thermostated solutions of [Fe(CN)₆]³⁻ and DL-methionine which also contained the necessary quantities of osmium(VIII), a catalyst, NaOH and NaClO₄ to maintain the required alkalinity and ionic strength, respectively. The temperature was maintained at 26 ± 0.1 °C. The course of reaction was followed by monitoring the decrease in absorbance of [Fe(CN)₆]³⁻ in 1 cm quartz cell of a Hitachi 150-20 spectrophotometer at its absorption maximum of 420 nm as a function of time. The application of Beer's law to [Fe(CN)₆]³⁻ at 420 nm had earlier been verified, giving ε = 1060 ± 50 dm³ mol⁻¹ cm⁻¹. An example run is given in Table III(1)(p.84) and Fig. III(1)(p.85). The first order rate constants, k.obs were evaluated by the plots of log [Fe(CN)₆]³⁻ versus time and were linear in almost all cases up to 85% completion of the reaction and the k.obs values were reproducibility to within ± 5%.

The effect of dissolved oxygen on the rate of the reaction was checked by preparing the reaction mixture and following the reaction in an atmosphere of N₂. No significant difference between the results obtained under N₂ and in presence of air was observed. In view of reaction carried out in basic medium, the effect of carbonate on the reaction was also studied. Added carbonate had no effect on the reaction rate. However, fresh solutions were used when conducting the experiment.
Table III(1)

Example run for the osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium at 26 °C.

\[
\begin{align*}
[DLM] &= 5.0 \times 10^{-3}; \\
[Fe(CN)_6]^{3-} &= 4.0 \times 10^{-4}; \\
[Os(VIII)] &= 1.0 \times 10^{-6}; \\
[OH^-] &= 0.20; \\
I &= 0.50 \text{ / mol dm}^{-3}.
\end{align*}
\]

<table>
<thead>
<tr>
<th>Time (min.)</th>
<th>Optical density (420 nm)</th>
<th>([Fe(CN)_6]^{3-} \times 10^4) (mol dm(^{-3}))</th>
</tr>
</thead>
<tbody>
<tr>
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</tr>
<tr>
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</tr>
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</tr>
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<td>12</td>
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</tbody>
</table>
Example run for the osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium at 420 nm.

\[
[DLM] = 5.0 \times 10^{-3}; \quad [\text{Fe(CN)}_6]^{3-} = 4.0 \times 10^{-4};
\]

\[
[\text{OH}^-] = 0.20; \quad I = 0.50 / \text{mol dm}^{-3}.
\]

(Conditions as in Table III(1)(p.84))
Stoichiometry and Product Analysis

Different sets of reaction mixtures containing different concentrations of DL-methionine and \([\text{Fe(CN)}_6]^{3-}\) at constant concentrations of osmium(VIII), alkali and ionic strength were kept for ca. 6 hours at 26 ± 0.1 °C in an inert atmosphere in a closed vessel. When \([\text{Fe(CN)}_6]^{3-}\) was higher than [DL-methionine], the unreacted \([\text{Fe(CN)}_6]^{3-}\) was found by measuring the absorbance at 420 nm spectrophotometrically. The results indicated that two moles of \([\text{Fe(CN)}_6]^{3-}\) consumed by one mole of DL-methionine as in equation (1).

\[
\text{OH} \quad \text{S} \quad \text{CH}_3 + 2[\text{Fe(CN)}_6]^{3-} + 2\text{OH}^- \xrightarrow{\text{Os(VIII)}} \text{OH} \quad \text{S} \quad \text{CH}_3 + 2[\text{Fe(CN)}_6]^{4-} + \text{H}_2\text{O} \quad (1)
\]

The main reaction product was identified as DL-methionine sulfoxide by spot test\(^9\) and it is also confirmed as below. The reaction mixture was allowed to stand for a few hours. Then sodium bicarbonate was added and stirred vigorously, followed by a drop wise addition of benzoyl chloride solution. The precipitate \(N\)-benzoyl methionine sulfoxide was confirmed\(^10\) by its m.p. 183 °C. The procedure is similar\(^11\) to the one employed in the oxidation of L-methionine by aqueous Cr(VI). The product is separated\(^12\) by adding acetone-ethanol mixture (1:1) to the reaction mixture at pH 4 resulted in the precipitate. The yield was found to be of about 90% for most the different sets of reaction mixtures. This also confirmed by
its m.p. (238 °C). The spectra of I.R. showed that the stretching for -NH₂ and -COOH were remained same. However, the new band for >S=O is appeared at 1032 cm⁻¹ for the product. No change in the NMR signals was observed. The reaction products do not undergo further oxidation to sulfone under the present kinetic conditions. This was confirmed by the test for sulfone⁹.

RESULTS

Reaction order

The order with respect to [DL-methionine], [Os(VIII)] and [OH⁻] were found by log \( k_\text{obs} \) versus log concentration plots.

Effect of [Hexacyanoferrate(III)]

The concentration of \([\text{Fe(CN)}_6]^{3-}\) was varied in the range, 8.0 \( \times 10^{-5} \) to 8.0 \( \times 10^{-4} \) mol dm⁻³ at fixed [DL-methionine], [Os(VIII)], [OH⁻] and ionic strength. The non-variation in the pseudo-first order rate constants at various concentrations of \([\text{Fe(CN)}_6]^{3-}\) indicates the order in \([\text{Fe(CN)}_6]^{3-}\) as unity (Table III(2)(p.88)). This was also confirmed from the linearity of plots of log (O.D) versus time \( (r>0.9994, \ S<0.026) \) (Fig. III(2)(p.89)) up to 85% completion of the reaction.

Effect of [DL-methionine]

The substrate, DL-methionine was varied in the range of 1.0 \( \times 10^{-3} \) to 1.0 \( \times 10^{-2} \) mol dm⁻³ at 26 °C keeping all other reactants concentrations constant (Table III(2)(p.88)). The \( k_\text{obs} \) values were constant with increase in concentration of DL-methionine indicating a zero order dependence on [DL-methionine].
Table III(2)

Effect of variation of $[\text{Fe(CN)}_6^{3-}]$ and [DL-Methionine] on osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium by $[\text{Fe(CN)}_6]^{3-}$ at 26 °C.

$[\text{OH}^-] = 0.20$;  
$[\text{Os(VIII)}] = 1.0 \times 10^{-6}$;  
$I = 0.50 \text{ mol dm}^{-3}$.

<table>
<thead>
<tr>
<th>$[\text{Fe(CN)}_6^{3-}] \times 10^4$ (mol dm$^{-3}$)</th>
<th>[DLM] $\times 10^3$ (mol dm$^{-3}$)</th>
<th>$k_{\text{obs}} \times 10^3$ (s$^{-1}$)</th>
<th>Found</th>
<th>Calculated</th>
</tr>
</thead>
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<td>1.39</td>
</tr>
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<td>10</td>
<td>1.35</td>
<td></td>
<td>1.39</td>
</tr>
</tbody>
</table>
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**Figure III(2)**

First order plots of osmium(VIII) catalyzed oxidation of DL-Methionine by hexacyanoferrate(III) at various [Fe(CN)$_6$]$^{3-}$ in aqueous alkaline medium at 26 °C at 420 nm.

(Conditions as in Table II(2)(p.88))

[Fe(CN)$_6$]$^{3-}$ = (1) 0.8 x 10$^{-4}$, (2) 1.0 x 10$^{-4}$, (3) 2.0 x 10$^{-4}$, (4) 4.0 x 10$^{-4}$, (5) 6.0 x 10$^{-4}$ and (6) 8.0 x 10$^{-4}$/mol dm$^{-3}$.
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Effect of [Alkali]

The effect of [alkali] on the rate of reaction was studied at constant concentrations of DL-methionine, $[\text{Fe(CN)}_6]^{3-}$ and ionic strength at 0.50 mol dm$^{-3}$ (Table III(3)(p.91). The rate constants increased with increase in [alkali] and the order was found to be $\approx 0.4$ (Fig. III(3)(p.92). Osmium(VIII) is known to be existed as $[\text{OsO}_3(\text{OH})_3]^\cdot$ in alkaline medium. Depending on the $[\text{OH}^-]$ used, the osmium(VIII) species changes as shown below.

$$\text{OsO}_3(\text{OH})_3^\cdot + \text{OH}^- \rightleftharpoons \text{OsO}_4(\text{OH})_2^{2+} + \text{H}_2\text{O} \quad K_1$$

$$\text{OsO}_4(\text{OH})_2^{2+} + \text{OH}^- \rightleftharpoons \text{OsO}_3(\text{OH})_3^- + \text{H}_2\text{O} \quad K_2$$

The total osmium(VIII) concentration, $[\text{Os(VIII)}]_T$ is the sum of $[\text{OsO}_3(\text{OH})_3]^\cdot$, $[\text{OsO}_4(\text{OH})_2]^{2-}$ and $[\text{OsO}_3(\text{OH})_3^-]$ and each of these are calculated from the known equilibria constants $K_1$ and $K_2$ (Table III(4)(p.93)). The results of such calculations are utilized to draw Fig. III(4)(.94) and it is seen that, the concentrations of different species, the variation of $[\text{OsO}_3(\text{OH})_3^-]$ with alkali shows any parallelism with the variation of rate constants with [alkali].

Effect of [Catalyst]

The concentration of osmium(VIII) was varied in the range, $5.0 \times 10^{-7}$ to $5.0 \times 10^{-6}$ mol dm$^{-3}$ at fixed $[\text{Fe(CN)}_6]^{3-}$, [DL-methionine], $[\text{OH}^-]$ and ionic strength (Table III(3)(p.91). and order was found to be unity (Fig. III(3)(p.92).

Effect of Ionic Strength and Solvent Polarity

The effect of ionic strength was studied by varying the NaClO$_4$ concentration in the reaction medium from 0.5 to 1.5 mol dm$^{-3}$ at constant $[\text{Fe(CN)}_6]^{3-}$, [DLM],
### Table III(3)

Effect of variation of \([\text{Os(VIII)}]\) and \([\text{OH}^-]\) on osmium(VIII) catalyzed oxidation of DL-methionine by \([\text{Fe(CN)}_6^{3-}]\) in aqueous alkaline medium at 26 °C.

\[ [\text{Fe(CN)}_6^{3-}] = 4.0 \times 10^{-4}, \quad [\text{DLM}] = 5.0 \times 10^{-3}, \]
\[ I = 0.50 \text{ mol dm}^{-3}. \]

<table>
<thead>
<tr>
<th>([\text{Os(VIII)}] \times 10^6) (mol dm(^{-3}))</th>
<th>([\text{OH}^-]) (mol dm(^{-3}))</th>
<th>(k_{\text{obs}} \times 10^3) (s(^{-1}))</th>
<th>Found</th>
<th>Calculated</th>
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</tr>
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<td>1.04</td>
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<td></td>
</tr>
<tr>
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</tr>
<tr>
<td>1.0</td>
<td>0.50</td>
<td>1.82</td>
<td>1.76</td>
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</tr>
</tbody>
</table>
Figure III(3)

Order in [OH\(^-\)] and [Os(VIII)] on the osmium(VIII) catalyzed oxidation of DL-Methionine by hexacyanoferrate(III) in aqueous alkaline medium at 26 °C.

(Conditions as in Table III(3)(p.91)
## Table III(4)

Variation of concentration of Different Os(VIII) species at various [OH'] at 26 °C.

<table>
<thead>
<tr>
<th>[OH'] (mol dm⁻³)</th>
<th>α₀</th>
<th>α₁</th>
<th>α₂</th>
<th>k_{obs} x 10^3 (s⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.05</td>
<td>0.3834</td>
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<td>0.70</td>
</tr>
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<td>0.08</td>
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<td>0.4843</td>
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<td>0.7112</td>
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<td>0.50</td>
<td>0.01859</td>
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<td>1.82</td>
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</table>

α₀, α₁ and α₂ are the fractions of concentrations of [OsO₃(OH)₃]⁻, [OsO₄(OH)₂]²⁻ and [OsO₄(OH)]³⁻ to [Os(VIII)]₇ respectively and are calculated as shown in equations (2) and (3).
**Figure III(4)**

Effect of [OH'] on different species of osmium(VIII) with $k_{obs}$ at 26 °C.

(Data from Table III(4)(p.93))

\[ \alpha_0, \alpha_1 \text{ and } \alpha_2 \text{ are the fractions of concentrations of OsO}_3(\text{OH})^3^-, \text{OsO}_4(\text{OH})_2^{2-} \text{ and OsO}_5(\text{OH})^3^- \text{ to } [\text{Os(VIII)}]_T \text{ respectively.} \]
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[Os(VIII)] and [OH'] (Table III(5)(p.96). It was found that the rate constants increased with increase in concentration of NaClO₄ and the plot of log \( k_{\text{obs}} \) versus \( \sqrt{I} \) was linear with positive slope (Fig. III(5)(p.97), \( r = 0.9989, s = 0.031 \)). The dielectric constant (D) effect was studied by varying the t-butyl alcohol-water content in the reaction mixture with all other conditions being constant. The dielectric constant were computed from the values of pure liquids\(^{13} \) as in Chapter II(p.62). Earlier it was found that there was no reaction of the solvent with the oxidant or catalyst under the experimental conditions used. The \( k_{\text{obs}} \) values increase with decrease in the dielectric constant of the medium (Table III(6)(p.98)). Hence, the graph of log \( k_{\text{obs}} \) versus \( 1/D \) was found to be linear with positive slope (Fig. III(5)(p.97)).

**Effect of Initially Added Products**

The initially added products, [Fe(CN)₆]⁴⁻ and [sulphoxide] were varied in the range of 2.0 x 10⁻⁵ to 2.0 x 10⁻⁴. (Table III(7)(p.99)) and found that they did not show any significant effect on the rate of the reaction.

**Polymerization study**

To test the intervention of free radicals, the reaction mixture was mixed with acrylonitrile monomer and kept for 24 hours under nitrogen atmosphere. On dilution with methanol, white precipitate of polymer was formed, indicating the presence of intervention of free radicals in the reaction. The blank experiment of either [Fe(CN)₆]³⁻, DL-methionine alone or mixture of catalyst and oxidant with acrylonitrile did not induce polymerization under the same condition as those induce with reaction mixture. Initially added acrylonitrile decreases the rate also indicating the free radical intervention, which is the case as in earlier work\(^{14} \).
### Table III(5)

Effect of variation of ionic strength (I) on the osmium(VIII) catalyzed oxidation of DL-Methionine by hexacyanoferrate(III) in aqueous alkaline medium at 26 °C.

\[
[DLM] = 5.0 \times 10^{-3}; \quad [\text{Fe(CN)}_6^{3-}] = 4.0 \times 10^{-4};
\]

\[
[\text{OH}^-] = 0.20; \quad [\text{Os(VIII)}] = 1.0 \times 10^{-6} / \text{mol dm}^{-3}
\]

<table>
<thead>
<tr>
<th>I (mol dm(^{-3}))</th>
<th>(\sqrt{I}) (mol dm(^{-3}))</th>
<th>(k_{\text{obs}} \times 10^3) (s(^{-1}))</th>
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Figure III(5)

Effect of ionic strength (I) and dielectric constant (D) on osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium at 26 °C.

(Conditions as in Table III(5)(p.96) and Table III(6)(p.98))
Table III(6)

Effect of variation dielectric constant (D) on the osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium at 26 °C.

\[
[DLM] = 5.0 \times 10^{-3}; \quad [\text{Fe(CN)}_6^{3-}] = 4.0 \times 10^{-4};
\]

\[
[\text{OH}^-] = 0.20; \quad [\text{Os(VIII)}] = 1.0 \times 10^{-6};
\]

\[I = 0.50 / \text{mol dm}^{-3}\]

<table>
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<tr>
<th>Water (%)</th>
<th>t-butyl alcohol-water (%) v/v</th>
<th>D</th>
<th>(k_{\text{obs}} \times 10^3) (s(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>100</td>
<td>0</td>
<td>78.50</td>
<td>1.37</td>
</tr>
<tr>
<td>95</td>
<td>5</td>
<td>75.12</td>
<td>1.43</td>
</tr>
<tr>
<td>90</td>
<td>10</td>
<td>71.74</td>
<td>1.55</td>
</tr>
<tr>
<td>85</td>
<td>15</td>
<td>68.36</td>
<td>1.65</td>
</tr>
<tr>
<td>80</td>
<td>20</td>
<td>64.98</td>
<td>1.82</td>
</tr>
</tbody>
</table>
### Table III(7)

Effect of the product, $[\text{Fe(CN)}_6]^{3+}$ and DL-methionine sulfoxide on the osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium at 26 °C.

$$[\text{DLM}] = 5.0 \times 10^{-3};$$

$$[\text{OH}^-] = 0.20;$$

$$[\text{Os(VIII)}] = 1.0 \times 10^{-6};$$

$$I = 0.50 / \text{mol dm}^{-3}.$$

<table>
<thead>
<tr>
<th>$[\text{Fe(CN)}_6]^{3+} / [\text{DLMSO}] \times 10^4$ (mol dm$^{-3}$)</th>
<th>$k_{\text{obs}} \times 10^3$ s$^{-1}$</th>
<th>[Fe(CN)$_6$]$^{3+}$</th>
<th>[DLMSO]</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.2</td>
<td>1.30</td>
<td>1.31</td>
<td></td>
</tr>
<tr>
<td>0.4</td>
<td>1.30</td>
<td>1.32</td>
<td></td>
</tr>
<tr>
<td>0.6</td>
<td>1.29</td>
<td>1.36</td>
<td></td>
</tr>
<tr>
<td>0.8</td>
<td>1.32</td>
<td>1.31</td>
<td></td>
</tr>
<tr>
<td>1.0</td>
<td>1.37</td>
<td>1.32</td>
<td></td>
</tr>
<tr>
<td>2.0</td>
<td>1.38</td>
<td>1.34</td>
<td></td>
</tr>
</tbody>
</table>
Effect of Temperature

The rate of reaction was measured at different temperatures under varying [OH'] at constant concentrations of oxidant, reductant, catalyst and ionic strength and the rate constants, k of slow step of Scheme 1 were obtained from intercepts of the plots of 1/k_{obs} versus 1/ [OH'] (Fig. III(6)(p.101)) (r>0.9998, S≤0.0152). The rate of reaction increased with the increase in temperature. The activation parameters corresponding to these constants for a slow step of reaction (Scheme I) were evaluated from the plot of log k versus 1/T (r>0.9989, S≤0.0135) (Table III(8)(p.102)).

DISCUSSION

Osmium(VIII) is known to form different complexes with OH' in basic media as shown in equation (2) and (3) with equilibrium constants of K_1 and K_2 having the values of 24 and 6.8 dm^3 mol\(^{-1}\) respectively\(^{15}\).

\[
\text{OsO}_3(\text{OH})_3^- + \text{OH}' \rightleftharpoons \text{OsO}_4(\text{OH})_2^{2-} + \text{H}_2\text{O} \quad K_1 \quad (2)
\]

\[
\text{OsO}_4(\text{OH})_2^{2-} + \text{OH}' \rightleftharpoons \text{OsO}_5(\text{OH})^{3-} + \text{H}_2\text{O} \quad K_2 \quad (3)
\]

Hence, the total [Osmium(VIII)] in alkaline medium is sum of [OsO_3(OH)_3^-], [OsO_4(OH)_2^{2-}] and [OsO_5(OH)^{3-}]. However, the [OsO_3(OH)_3^-] is existed in lower [OH'] and at [OH'] used in the present study, virtually all Os(VIII) is present as [OsO_4(OH)_2^{2-}] and [OsO_5(OH)^{3-}]. The fractions of total [Os(VIII)] species, [OsO_4(OH)_2^{2-}] and [OsO_5(OH)^{3-}] calculated using K_1 and K_2 reveal that the [OsO_5(OH)^{3-}] with different [OH'] are parallelism with [OH'] (Fig. III(4)(p.94))
Verification of rate law of osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium at different temperatures.

\[
[DLM] = 5.0 \times 10^{-3}; \quad [\text{Fe(CN)}_6^{3-}] = 4.0 \times 10^{-4};
\]

\[
[\text{Os(VIII)}] = 1.0 \times 10^{-6}; \quad I = 0.50 / \text{mol dm}^{-3}.
\]
### Table III(8)

**A:** Effect of temperature on the osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium.

<table>
<thead>
<tr>
<th>Temp. (K)</th>
<th>$k_{obs} \times 10^3$ (dm$^3$ mol$^{-1}$ s$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>299</td>
<td>1.30</td>
</tr>
<tr>
<td>304</td>
<td>1.62</td>
</tr>
<tr>
<td>309</td>
<td>1.96</td>
</tr>
<tr>
<td>314</td>
<td>2.43</td>
</tr>
</tbody>
</table>

**B:** Activation parameters for the osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium.

<table>
<thead>
<tr>
<th>Log $A$ (k J mol$^{-1}$)</th>
<th>$E_a$ (k J mol$^{-1}$)</th>
<th>$\Delta H^#$ (kJ mol$^{-1}$)</th>
<th>$\Delta S^#$ (J K$^{-1}$ mol$^{-1}$)</th>
<th>$\Delta G^#$ (kJ mol$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.66 ± 0.2</td>
<td>31.6 ± 0.2</td>
<td>29.2 ± 0.2</td>
<td>-201 ± 10</td>
<td>89 ± 4</td>
</tr>
</tbody>
</table>

**C:** Thermodynamic parameters for the osmium(VIII) catalyzed oxidation of DL-methionine by hexacyanoferrate(III) in aqueous alkaline medium (step 1 of Scheme 1 at 299K).

<table>
<thead>
<tr>
<th>$\Delta H$ (kJ mol$^{-1}$)</th>
<th>$\Delta S$ (J K$^{-1}$ mol$^{-1}$)</th>
<th>$\Delta G$ (kJ mol$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>8.7 ± 0.3</td>
<td>48 ± 2.0</td>
<td>-5.6 ± 0.2</td>
</tr>
</tbody>
</table>
Because of this reason and the fact that the $k_{obs}$ is a function of $[\text{OH}^-]$, the main active species of catalysis is likely to be $[\text{OsO}_4(\text{OH})]^3-$ and its formation in the equilibrium (3) is of importance in this study.

The first order each in catalyst and oxidant, fractional order in alkali and zero order in DL-methionine accommodate the mechanism as given in Scheme 1, where osmium(VIII) forms its hydroxide species in prior equilibrium step, interact with $[\text{Fe(CN)}_6]^{3-}$ in a slow step to yield a complex. This complex reacts with DL-methionine in a fast step to give DL-methionine free radical followed by formation of DL-methionine sulfoxide in the subsequent fast step.

$$[\text{OsO}_4(\text{OH})_2]^{2-} + \text{OH}^- \xrightleftharpoons{K_2} [\text{OsO}_4(\text{OH})]^3^- + \text{H}_2\text{O}$$

$$[\text{OsO}_4(\text{OH})]^3^- + [\text{Fe(CN)}_6]^{3-} \xrightarrow{k_{\text{slow}}} C_1 + \text{CN}^-$$

$$C_1 + \text{O} \xrightarrow{\text{fast}} \text{O}$$

$$+ [\text{Fe(CN)}_6]^{3-} + [\text{OsO}_4(\text{OH})]^3^-$$

$$[\text{Fe(CN)}_6]^{3-} + \text{CN}^- \xrightarrow{\text{fast}} [\text{Fe(CN)}_6]^{4-}$$
The complex (C<sub>1</sub>) might have the structure

$$\begin{array}{c}
\text{NC} \\
\text{N} \\
\text{O} \\
\text{C} \\
\text{O}
\end{array}$$

Though the DL-methionine contains the coordinating centers nitrogen and oxygen of -NH<sub>2</sub> and -COOH group as the coordinating centers and sulfur as the sites of attacking by the oxidant or a catalyst, the ‘S’ is oxidized to sulfoxide as per the sequence<sup>16</sup> of the reaction taking place in sulfoxides is R-S-R > R-SO-R > R-SO<sub>2</sub>-R. The sulfide to sulfoxide step is fast as observed in the present study compared to the sulfoxide to sulfone step.

In the present study the sulfoxide was found to be the main oxidation product which supports the above facts. Thus, the oxidation at the sites of nitrogen and oxygen is ruled out and it is an example for direct electron transfer reaction.

The intermediate complex (C<sub>1</sub>) due to interaction between osmium(VIII) and [Fe(CN)<sub>6</sub>]<sup>3-</sup> in the rate determining step is written as reported<sup>17</sup> by earlier
workers which justifies the unit order each in oxidant and catalyst and zero order in substrate. The formation of DL-methionine free radical in the fast step is in accordance with the experimental result of retardation of rates by the added radical scavenger like acrylonitrile and formation of copious precipitate on diluting with methanol. The scheme 1 is written in accordance with experimentally observed facts. Generally, in Os(VIII) catalyzed reactions, Os(VIII) oxidizes the substrate followed by reoxidation of Os(VI) with oxidant in alkali. If that is the case, the order with respect to substrate would be unity or fraction and there could not be an intervention of free radicals. However, in the present study the above facts could be ruled out as order with respect to substrate is zero and reaction occurring through intervention of free radical. The another factor of reoxidation of Os(VI) by [Fe(CN)₆]³⁻ can be ruled out as their redox potentials do not permit the reaction. This also confirmed from their direct reaction. The another factor of noncatalytic influence of [Fe(CN)₆]⁴⁻ in the present study support the mechanism written in Scheme 1. From the Scheme 1, the rate law can be derived as below,

\[
\frac{-d[Fe(CN)₆^{3-}]_T}{dt} = \text{Rate} = k[Fe(CN)_₆]^{3-}[OsO₅(OH)]^{3-} \\
= K_2k[Fe(CN)_₆]^{3-}[OsO₄(OH)_₂]^{2-}[OH^{-}]_f \quad (4)
\]

[Os(VIII)]ₖ is equal to the sum of the concentrations of OsO₄(OH)²⁻ and OsO₅(OH)³⁻.

i.e. \[\text{[Os(VIII)]}_T = [OsO₄(OH)_₂]^{2-} + [OsO₅(OH)]^{3-}\]

\[= [OsO₄(OH)_₂]^{2-} + K_2[OsO₄(OH)_₂]^{2-}[OH^{-}]_f \]

\[= [OsO₄(OH)_₂]^{2-}\{1 + K_2[OH^{-}]_f\}\]
Therefore, from equation (5) and (6), the equation (4) becomes,

\[
\text{Rate} = \frac{kK_2[\text{Fe(CN)}_6]^3[\text{Os(VIII)}][\text{OH}]}{1 + K_2[\text{OH}]} \cdot \frac{[\text{OH}]}{1 + K_2[\text{OsOs(OH)}_2]^2} \]  

\[ (7) \]

In view of low concentration of Os(VIII) used, the term \( (1 + K_2[\text{OsOs(OH)}_2]^2) \) tends to unity in the denominator. Therefore, the above equation becomes,

\[
\text{Rate} = \frac{kK_2[\text{Fe(CN)}_6]^3[\text{Os(VIII)}][\text{OH}]}{1 + K_2[\text{OH}]} \]  

\[ (8) \]

For the verification of rate law the subscripts ‘T’ and ‘f’ are omitted and hence, equation (7) becomes,

\[
\frac{\text{Rate}}{[\text{Fe(CN)}_6]^3} = k_{\text{obs}} = \frac{kK_2[\text{Os(VIII)}][\text{OH}]}{1 + K_2[\text{OH}]} \]  

\[ (8) \]

The mechanism shown in Scheme 1 and rate law (8) are verified by a plot of \( 1/k_{\text{obs}} \) versus \( 1/[\text{OH}] \) which should be linear and slope and intercept should lead
to the values of $K_2$ and $k$. Such a plot is shown in Fig. III(6)(p.101) with $K_2 = 9.6 \pm 0.4 \text{ dm}^3 \text{ mol}^{-1}$ ($K = 6.8 \text{ dm}^3 \text{ mol}^{-1}$ at 50 °C from earlier work\textsuperscript{18} and $k = 2.12 \times 10^3 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$. The values of $k$ and $K_2$ were further used to calculate rates for several experimental situations. Rate constants calculated in this way have been found to be comparable with experimentally measured rate constants (Table III(2)(p.88)) and (Table III(3)(p.91)) and there is generally a good agreement.

The effect of increasing ionic strength on the rate is also in the expected direction on the basis of the reaction between the negatively charged $\text{OsO}_3(\text{OH})^{3-}$ and $[\text{Fe(CN)}_6]^{3-}$ in the rate determining step. Ion pairing between $[\text{Fe(CN)}_6]^{3-}$ and $K^+$ is well known\textsuperscript{19}. Thus, ion pairing between $\text{Na}^+$ and $[\text{Fe(CN)}_6]^{3-}$ may be expected in the presence of NaOH and NaClO$_4$ which were used to maintain [alkali] and ionic strength respectively. Thus, the overall increase of $k_{\text{obs}}$ with increase in ionic strength (Table III(5)(p.96)).

The increase in rate with decreasing the dielectric constant of the media is contrary to the expected direction\textsuperscript{20} as the ionic species involved in the slow step. This may be due to the more solvation of activated complex at low dielectric constant media than at higher one where the reactants are more solvated. The hydroxyl ion concentration (Fig. III(6)(p.101)) was varied at four different temperatures; and from the slopes and intercepts of plots of $1/k_{\text{obs}}$ versus $1/[\text{OH}^-]$ the $K_2$ values were determined as 9.6 dm$^3$ mol$^{-1}$, 10.1 dm$^3$ mol$^{-1}$, 10.7 dm$^3$ mol$^{-1}$, 11.3 dm$^3$ mol$^{-1}$ at 26, 31, 36 and 41 °C respectively. The Van't Hoff's plot was made for the variation of $K_2$ with temperature (i.e. log $K_2$ versus $1/T$).
Chapter III

(Fig. III(7)(p.109)). The thermodynamic parameters like enthalpy change (ΔH) entropy change of the reaction (ΔS) and free energy change of reaction (ΔG) for the first step of Scheme 1 were calculated (Table III(8)(p.102)). And these values are compared with the values obtained for slow step of the reaction shows that these values mainly refer to the rate limiting step, supporting the fact that the reaction before the rate determining step are fairly slow and involves high activation energy 21.

A large negative value of ΔS (-201 kJ) suggests that the two ionic species combine in rate determining step to give an intermediate complex which is more ordered than the reactants 20,22. The higher rate constants of the slow step of the mechanism indicated that the oxidation presumably occurs by an inner-sphere mechanism. This conclusion was supported by earlier work 23. The unexpected low value of frequency factor (log A = 2.66) clearly indicates that the similar charged ions are interacting rather than DL-methionine with Os(VIII) or [Fe(CN)₆]³⁻ in a slow step which supports the mechanism as in Scheme 1.
Figure III(7)

Van't Hoff’s plot for the calculation of thermodynamic parameters at different temperatures.
FINDINGS

Uncatalyzed alkaline hexacyanoferrate(III) oxidation of DL-methionine in alkali occurs with a little rate, however, the same in presence of about 0.1 μ mole of osmium(VIII) under the identical condition, the rate of reaction multiplies in the order of more than a million times (half life period of unanalyzed reaction is 1,20,000 minutes where as Osmium(VIII) catalyzed under the same reaction condition is 5 minutes). This may be due to formation of intermediate compound of a mixture of hexacyanoferrate(III) which facilitates the transfer of electron from reductant to oxidant through a common bridging ligand ‘O’ of osmium(VIII) (vide probable structure of complex C1). The oxidation product of DL-methionine was found to be DL-methionine sulfoxide which is unlike to the formation of L-methionine from D-methionine in the metabolism via the formation of keto-acid followed by reamination. The role of [Fe(CN)6]4- is not observed as its catalytic effect might be masked in presence of a strong catalyst like Os(VIII). Thermodynamic parameters of first step of Scheme 1 supports the increase in concentration of OsO4(OH)3+ with increase of [OH⁻] as ΔG has a very small negative value.
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