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Insight into D-galactose Oxidation: Kinetic and Mechanistic Analysis with Imidazolium Fluorochromate

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Abstract: In a 50% (v/v) aqueous acetic acid medium, a study on the oxidation kinetics of D-galactose by imidazolium fluorochromate (IFC) was conducted. The study revealed unit-order dependence on [D-galactose], [IFC], and [perchloric Acid]. The reaction was catalyzed by perchloric acid, and followed a 1:1 stoichiometric ratio. Adding sodium hyperchlorate (VII) salt did not affect the reaction. Polymerization of acrylonitrile was not observed in the oxidation process. The oxidation rates were retarded on adding Mn^{2+} ions in the mixture, and rates increased as the reaction medium's dielectric constant (D) decreased. The observed results follow the Amis and Kirkwood plots {log k_1 vs. (1/D) and ((D-1/2D+1))}. Several thermodynamic variables were determined by analyzing the kinetic data across a temperature range. Arabinose and formic acid were formed as the products of the reaction. A mechanistic path involving the formation of an ester followed by hydride ion transfer was proposed.

Keywords: Oxidation, Kinetics, Mechanism, D-galactose, Imidazolium Fluorochromate

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1. INTRODUCTION

Chromium (VI), in the form of chromate or dichromate, is highly toxic and soluble in water, but generally insoluble in most organic solvents (1). However, sugars and their derivatives can influence chromium chemistry due to their reducing properties (2). It was observed that oxidation process could convert potentially hazardous Cr (VI) into environmentally benign and non-toxic Cr (III)/Cr (IV) oxidation states (3). Therefore, various Cr (VI) complexes like pyridinium chlorochromate (4), tetraethylammonium chlorochromate (5), quinolinium chlorochromate (6), quinolinium fluorochromate (7), pyridinium fluorochromate (8), and tributylammonium chlorochromate (9) have been developed to oxidize some biologically important organic molecules, including sugars (10). D-galactose, a specific type of reducing sugar, plays an important role as a primary energy source for mammalian infants during their exclusive dependence on milk. The primary origin of galactose is lactose, found in dairy products, which undergoes hydrolysis to form glucose and galactose. Moreover, small amounts of galactose can be found in certain vegetables and fruits. Recent researches have suggested potential therapeutic applications for galactose, including its use in the treatment of diseases like Alzheimer's and nephrotic syndrome (11–15). Given its crucial role in neonates' metabolism and therapeutic benefits, a study on oxidation kinetics was performed. To control selectivity and over-oxidation of products, imidazolium fluorochromate (IFC) (16,17), a recently developed Cr (VI) complex was used as an oxidant in the reaction. Previous literature studies illustrated that oxidation study of D-galactose with IFC has not been reported yet (17,18). Considering this, the kinetic and mechanistic characteristics of the oxidation of D-galactose with IFC oxidant are described here.

2. EXPERIMENTAL SECTION

2.1 Materials and methods

IFC oxidant has been prepared by the reported method (17). The purity of oxidant was confirmed by using dichromatic method. Commercially provided D-galactose was employed as the substrate. To ensure consistency, the D-galactose solution was always prepared fresh in a 50% (v/v) aqueous acetic acid solvent medium. Sodium hyperchlorate (VII) salt was added to maintain an unaltered ionic strength throughout the reaction mixture. Perchloric acid was used as a source of active H^+ ions in the reaction. All TLC experiments were performed on an activated cellulose MN 300 G layer plate, as stated in the published method (19). All other chemicals and solvents were used without any further purification.

2.2 Kinetic analysis

The kinetics were studied under pseudo-first-order conditions when [D-galactose] is in excess over [IFC]. A 50% (v/v) aqueous acetic acid served as the solvent, unless specified differently. The reaction advancement was tracked by assessing absorbance at 350 nm through a visible spectrophotometer, reaching 70-80% completion. Pseudo-first order velocity constants (denoted as k_1) were evaluated from the slope of the linear plot between $\log_e (A_0/A_t)$ and time (t) {Here, A_0 represents absorbance value when the reaction commenced, and A_t signifies the absorbance at any specific time}. Second-order rate constants (k_2) were computed using the given expression (i):

$$k_2 = \frac{k_1}{[\text{D-galactose}]} \qquad (i)$$

2.3 Stoichiometry

It was established through a mixture where [D-galactose] was in excess of [IFC], kept in darkness at 303 K to attain a complete reaction. The remaining Cr (VI) was determined by dichromatic approach. A 1:1 stoichiometric ratio was observed in the reaction, consistent with the stated expression (ii):

$$\underset{\text{D-Galactose}}{C_6H_{12}O_6} + \underset{\text{IFC}}{(IH)OCrO_2F} \xrightarrow{H^+} \underset{\text{D-Arabinose}}{C_5H_{10}O_5} + HCOOH + \underset{\text{Cr(IV)}}{[(IH)OCr(OH)F)^+]} \qquad (ii)$$

2.4 Product analysis

For product assessment, a 1:1 molar ratio of D-galactose and IFC was prepared and equilibrated for 24 hours. The products were separated using trichloromethane ($CHCl_3$) and dried over anhydrous $MgSO_4$ (20). The oxidation product was identified as D-arabinose by comparing its R_f value with authentic sugar samples (Table 1). The formation of a phenylhydrazone derivative also supports the formation of D-arabinose. Further, formic acid was detected as a side product through spot test analysis (20–22).

Table 1: The R_f values of standard sugars in comparison with product

Sugars	R_f (Std. sugar)	R_f (Product)
Galactose	0.34	-
Mannose	0.47	-
Fructose	0.46	-
Arabinose	**0.52**	**0.52**
Xylose	0.68	-
Glucose	0.42	-
Ribose	0.71	-

3. RESULTS AND DISCUSSION

3.1 Effect on varying [oxidant]

At constant [D-galactose], [$HClO_4$], ionic strength, and temperature, the reaction was examined at different [IFC], and pseudo-first-order rate constants (k_1) were computed. It was observed that k_1 remained constant irrespective of [IFC], which implies a first-order nature of the reaction concerning [IFC] (Table 2). The same conclusion is also noticeable from a linear plot between ($\log_e (A_0/A_t)$) and time (Figure 1).

3.2 Effect of varying [substrate]

The rate constants (k_1) were calculated at constant [IFC], [$HClO_4$], ionic strength, and temperature. The observed k_1 values were elevated and demonstrated a direct proportionality with [D-galactose] (Figure 2(a)), indicating a first-order relationship between substrate and the reaction rate (Table 2). Further, the slope of the plot between ($\log k_1$) and (log [D-galactose]) is 0.9875, which also justifies the observed result (Figure 2(b)).

3.3 Effect of varying [perchloric acid]

The k_1 was evaluated at different [$HClO_4$], keeping all other reaction conditions the same. The study revealed that the rates increased with the base to the power of one in [$HClO_4$], which suggests the reaction is first order concerning [$HClO_4$], as illustrated in Figure 3(a). In addition, the gradient of the observed linear plot between ($\log k_1$) and ($\log [HClO_4]$) is 1.0024, corroborating the findings (Figure 3(b)).

Table 2: Rate constants (k_1 & k_2) for oxidation process of D-galactose by IFC at 303 K.

[IFC] x 10³ M	**[D-galactose] x 10² M**	**[$HClO_4$] M**	**k_1 x 10⁴ (s^{-1})**	**k_2 x 10² (M^{-1} x s^{-1})**	**R^2**
0.66	2.66	0.89	7.46		0.9911
1.33	2.66	0.89	7.69	2.89	0.9942
2.66	2.66	0.89	7.57		0.9899
4.00	2.66	0.89	7.76		0.9927
1.33	1.33	0.89	3.63	2.73	0.9781
1.33	4.00	0.89	11.05	2.76	0.9942
1.33	5.33	0.89	14.76	2.77	0.9958
1.33	6.66	0.89	18.27	2.74	0.9967
1.33	8.00	0.89	22.21	2.78	0.9844
1.33	2.66	0.668	5.82		0.9901
1.33	2.66	1.113	9.85		0.9959
1.33	2.66	1.316	11.38		0.9983
1.33	2.66	1.558	13.87		0.9979
1.33	2.66	1.781	15.29		0.9936
1.33	2.66	2.004	17.48		0.9966

3.4 Effect of adding salt

The reaction was examined across various $NaClO_4$ concentrations, from 1.33 x 10^{-2} to 4.66 x 10^{-2} M. Interestingly, the reaction rates remain constant, indicating the involvement of primary and secondary salt effects, potentially nullifying each other. A literature study also supports the observed findings (23–25).

3.5 Effect of incorporating a radical precursor

To understand the reaction pathway, acrylonitrile, a radical forming species, was introduced into the mixture. The absence of acrylonitrile polymerization in the reaction indicates that the formation of free radicals was unlikely during the reaction.

3.6 Effect of adding Mn^{2+} ions

Mn^{2+} ions were introduced into the reaction mixture to investigate the potential involvement of Cr^{4+} as an intermediate in the oxidation process. A study previously recognized Mn^{2+} as a reliable agent for trapping Cr^{4+} intermediates (26). Increasing [Mn^{2+}] led to a decrease in the reaction rates (Table 3), indicating the participation of Cr^{4+} with Mn^{2+}, as illustrated in expression (iii):

$$Cr^{4+} + Mn^{2+} \rightarrow Cr^{3+} + Mn^{3+} \qquad \text{(iii)}$$

Table 3: Mn^{2+} ion-based rate constants (k_1) for the D-galactose oxidation by IFC oxidant in a 50% (v/v) aqueous acetic acid medium at 303 K. [IFC] = 1.33 x 10^{-3} M, [D-galactose] = 2.66 x 10^{-2} M, [$HClO_4$] = 0.89 M, [$NaClO_4$] = 0.13 M

[$MnSO_4$] x 10² M	**k_1 x 10⁴ (s^{-1})**	**R^2**
0.00	7.69	0.9942
2.66	7.45	0.9928
5.33	7.26	0.9969
10.66	6.92	0.9914
16.00	6.54	0.9918

3.7 Effect of varying solvent composition

Kinetics was analyzed in solvents with different acetic acid: water compositions to gain insight into the interaction between the reacting species. A decrease in rate was observed in solvents with high dielectric constants (Table 4).

Table 4: Solvent based rate constants (k_1) for the oxidation of D-galactose by IFC reagent at 303 K. [IFC] = 1.33 x 10^{-3} M, [D-galactose] = 2.66 x 10^{-2} M, [$HClO_4$] = 0.89 M, [$NaClO_4$] = 0.13 M

CH_3COOH: H_2O	**k_1 x 10^4 (s^{-1})**	**R^2**	**1000/D**	**D-1/2D+1**
40:60	5.92	0.9923	20	0.485
50:50	7.69	0.9942	24	0.482
60:40	10.82	0.9958	28	0.479
70:30	14.71	0.9903	36	0.473
80:20	22.36	0.9875	48	0.465

The positive gradient of the linear Amis plot (log k_1 and 1/D) and the linear Kirkwood plot (log k_1 and (D-1/2D+1)) suggest ion-dipole interactions in the reaction's suggested mechanistic pathway {Here, D denotes the dielectric constant of the reaction medium} (Figure 4(a) and 4(b)) (27,28).

3.8 Effect of varying temperatures

A temperature-dependent trend in second-order rate constants (k_2) was studied by recording kinetics over different temperature ranges (T). The k_2 values showed an increase with rising temperatures, as depicted in Table 5. The activation energy (E_a), followed by enthalpy of activation ($\Delta H^‡$) were determined from the linear plot between ($\log_e k_2$) and (1/T) (Figure 5) and expression (iv).

$$\Delta H^‡ = E_a - 2RT \qquad \text{(iv)}$$

Free energy and entropy of activation ($\Delta G^‡$ and $\Delta S^‡$) were computed using Eyring's equation for reaction rates (expression (v)) (24,25,29).

$$k_2 = K_t \frac{K_b T}{h} e^{-\frac{\Delta G^‡}{RT}} \qquad \text{(v)}$$

The observed negative value of entropy of activation ($\Delta S^‡$) indicates that the intermediate complex is more polar compared to the initial reactants. The overall decrease in entropy during complex formation is typically due to the loss of translational energy required to bring multiple reactants together at the transition state (23, 30, 31).

Table 5: Temperature-based rate constants (k_2) and thermodynamic parameters for the D-galactose oxidation by IFC oxidant in a 50% (v/v) aqueous acetic acid medium. [IFC] = 1.33 x 10^{-3} M, [D-galactose] = 2.66 x 10^{-2} M, [$HClO_4$] = 0.89 M, [$NaClO_4$] = 0.13 M

Temp (K)	**k_2 x 10^2 (M^{-1} x s^{-1})**	**E_a (kJ/mol)**	**$\log_e A$ (M^{-1} x s^{-1})**	**$\Delta G^‡$ (kJ/mol)**	**$\Delta H^‡$ (kJ/mol)**	**$-\Delta S^‡$ (J K^{-1} mol^{-1})**
293	1.49	56.11	18.82	81.99	51.24	104.94
303	2.89		18.73	83.20	51.07	106.03
313	5.78		18.71	84.22	50.91	106.42
323	11.26		18.71	85.21	50.74	106.71
		Mean:	18.74 ±0.03	83.66 ±0.69	50.99 ±0.11	106.03 ±0.39

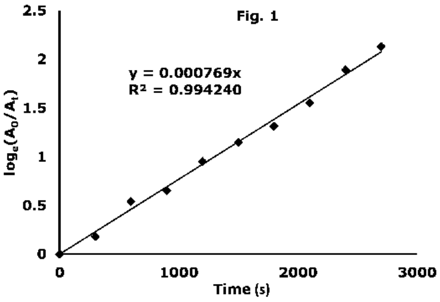


Figure 1: Plot between $\log_e (A_0/A_t)$ and Time (s) for oxidation of D-galactose by IFC oxidant in a 50% (v/v) aqueous acetic acid medium at 303 K. [IFC] = 1.33 x 10^{-3} M, [D-galactose] = 2.66 x 10^{-2} M, [$HClO_4$] = 0.89 M, [$NaClO_4$] = 0.13 M

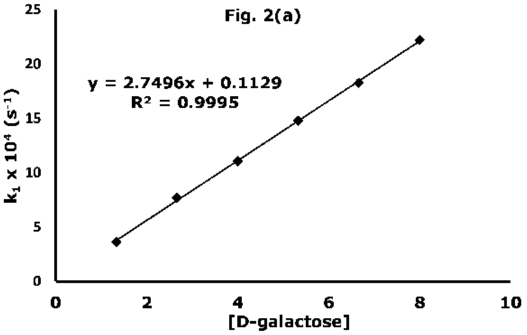


Figure. 2(a): Plot between (k_1) and ([D-galactose]), **2(b):** Plot between ($\log k_1 + 4$) and (log [D-galactose] + 2) for oxidation of D-galactose by IFC oxidant in a 50% (v/v) aqueous acetic acid medium at 303 K. [IFC] = 1.33 x 10^{-3} M, [$HClO_4$] = 0.89 M, [$NaClO_4$] = 0.13 M

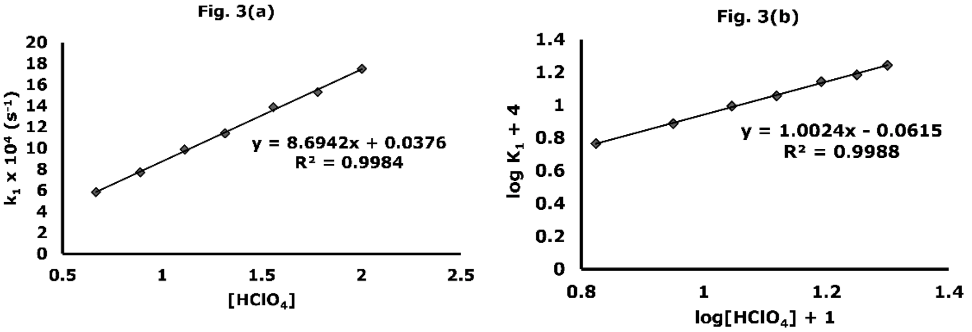


Figure 3(a): Plot between (k_1) and ([$HClO_4$]), **3(b):** Plot between ($\log k_1 + 4$) and (log [$HClO_4$] + 1) for oxidation of D-galactose by IFC oxidant in a 50% (v/v) aqueous acetic acid medium at 303 K. [IFC] = 1.33 x 10^{-3} M, [D-galactose] = 2.66 x 10^{-2} M, [$NaClO_4$] = 0.13 M

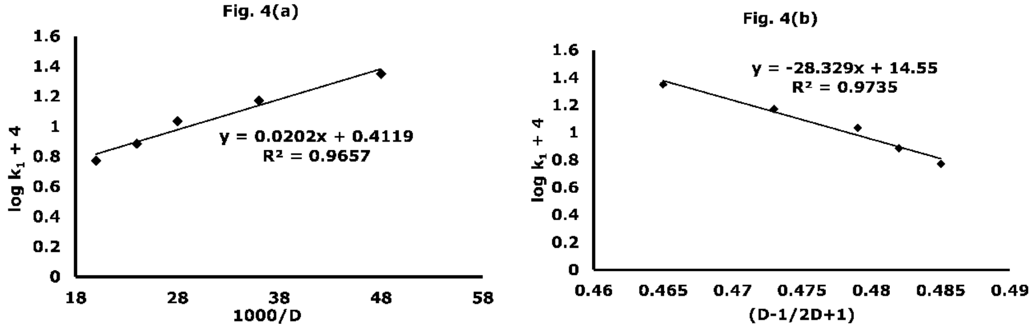


Figure 4 (a): Plot between ($\log k_1 + 4$) and (1000/D) (Amis plot), **4 (b):** Plot between ($\log k_1 + 4$) and (D-1/2D+1) (Kirkwood plot) for the oxidation of D-galactose by IFC oxidant at 303 K. [IFC] = 1.33 x 10^{-3} M, [D-galactose] = 2.66 x 10^{-2} M, [$HClO_4$] = 0.89 M, [$NaClO_4$] = 0.13 M

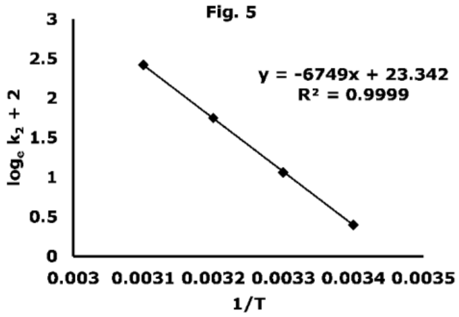


Figure 5: Plot between ($\log_e k_2 + 2$) and (1/T) for D-galactose oxidation by IFC oxidant in a 50% (v/v) aqueous acetic acid medium. [IFC] = 1.33×10^{-3} M, [D-galactose] = 2.66×10^{-2} M, [$HClO_4$] = 0.89 M, [$NaClO_4$] = 0.13 M

3.9 Mechanism of the reaction

In an acidic aqueous condition, D-galactose primarily adopts a cyclic hemiacetal, resulting in mixture of α- and β-pyranose ring forms that constantly interconvert with the rare open-chain structure (Scheme 1) (32). Among these, the prevalent form is β-D-galactopyranose (Table 6).

Table 6: Percentage Composition of different galactose configurations (33)

Aldose	[α-Pyranose] (%)	[β-pyranose] (%)	Open chain (%)
Galactose	~35	~65	0.085

Reaction involving monosaccharides proceeds through the acyclic configuration, probably feasible by converting the chain form to open-chain form through a less favorable equilibrium step (34). A comparative kinetic study on the reactivity of several sugars with Cr(VI) revealed that their reactivity increased proportionally with an increasing amount of open-chain configuration of the sugar (33,35). Some reports also suggest that cyclic pyranose forms have increased exposure to interact with Cr(VI), which can actively participate in oxidation processes (36–38). Therefore, it is rational to infer that the rate constant (k_1) encompasses the combined influence of the cyclic and acyclic forms, both of which persist in a state of dynamic equilibrium.

In the presence of acid, IFC (A) is protonated; species (B) is formed by selectively capturing the released H^+ ion from the dissociation of $HClO_4$ (Scheme 2). Additionally, the first-order reliance on [$HClO_4$] and the catalytic impact of perchloric acid on reaction rates signifies that the oxidation process exclusively takes place under acidic conditions, and the equilibrium constant for the protonation step is low.

Notably, no fractional order kinetics dependence on [Substrate] and the absence of spectrophotometric proof indicate the absence of the proposed complex. Nonetheless, these findings do not conclusively dismiss the possibility of the proposed complex. It was hypothesized that the rate constants governing the complex formation step are very small, explaining the absence of observable kinetic and spectrophotometric evidence (34, 39–41). Previous studies on the oxidation process of various organic compounds using Cr(VI)-based oxidants have reported an ester complex formation mechanism (40–43). Considering all these facts into account, it is postulated that the charged version of IFC interacts with the cyclic β-form of galactose and an ester-like complex is formed. Subsequently, this ester complex undergoes a hydride ion transfer in the rate-limiting step of the reaction, followed by C_1-C_2 bond cleavage, resulting in the formation of the products outlined in Scheme 3.

3.10 Rate law

$$(IH)OCrO_2F + H^+ \overset{K_1}{\Leftrightarrow} [(IH)OCr(O)(OH)F)^+] \qquad (vi)$$

$$\text{D-galactose} + [(IH)OCr(O)(OH)F)^+] \overset{K_2}{\Leftrightarrow} \text{Ester-complex} + H_2O \qquad (vii)$$

$$\text{Ester-complex} \xrightarrow{k,\text{slow}} \text{Arabinose} + \text{Formic acid} + [(IH)OCr(OH)F)^+] \qquad (viii)$$

The rate equation for the proposed mechanism was formulated by using expression (viii), which serves as the rate-determining step of the reaction.

$$\frac{-d[IFC]}{dt} = k[\text{ester-complex}] \qquad (ix)$$

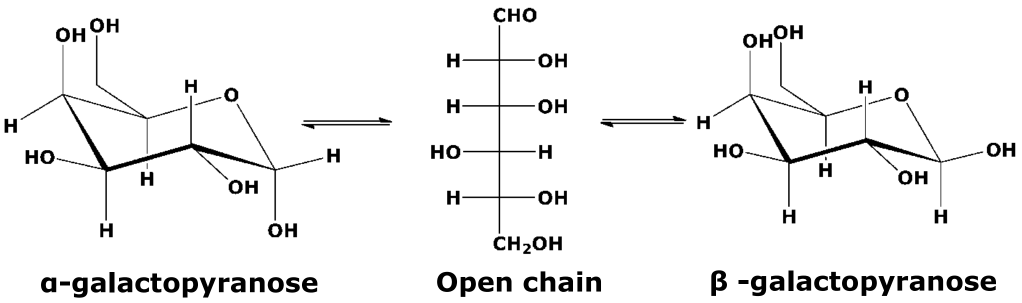
On applying the approximation condition in expression (vii) and using expression (vi), expression (ix) could be written as,

$$\frac{-d[IFC]}{dt} = \frac{kK_1K_2[\text{D-galactose}][IFC]_t[H^+]}{(1+K_1K_2[\text{D-galactose}][H^+])} \qquad (x)$$

In expression (x), $[IFC]_t$ = [IFC] + [ester-complex]. Taking into account the previously discussed small values of K_1 and K_2, it is logical to assume that 1>>>(K_1K_2[D-galactose][H^+]) and $[IFC]_t$ = [IFC]. This assumption gives rise to the following rate law (expression (xi)), explaining the experimental results observed.

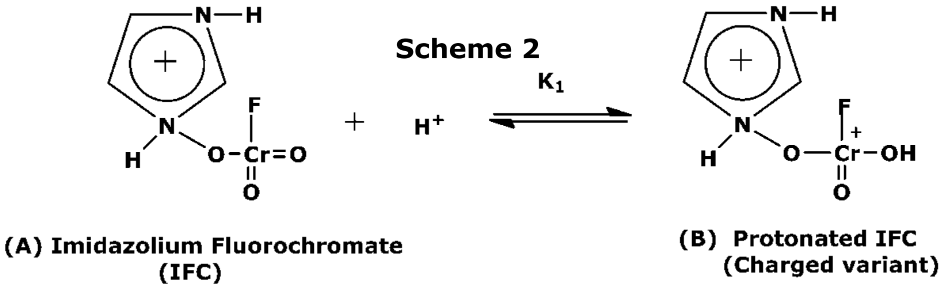
$$\frac{-d[IFC]}{dt} = kK_1K_2[\text{D-galactose}][IFC][H^+] \qquad (xi)$$

Scheme 1



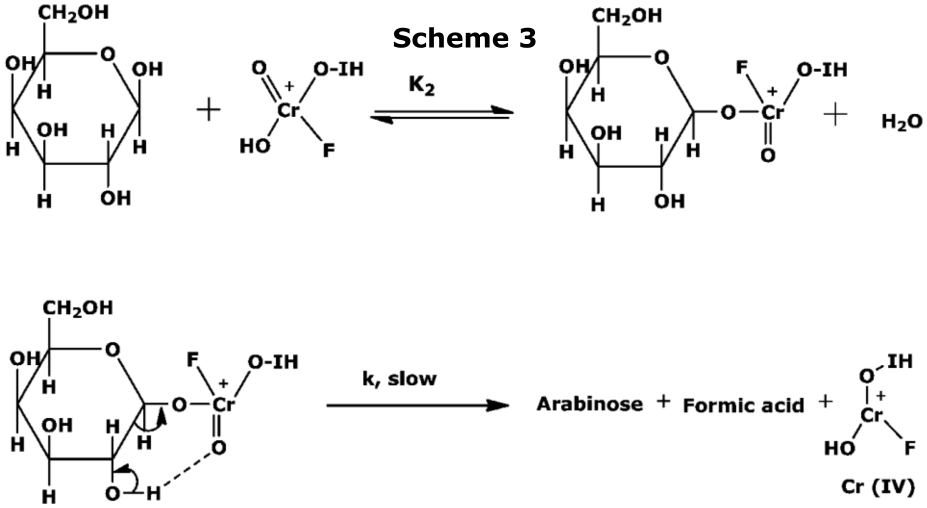
Scheme 1: Interconversion of α-galactopyranose to β-galactopyranose via open chain form

Scheme 2



Scheme 2: Structure of IFC and its Protonated form

Scheme 3



Scheme 3: Reaction mechanism

4. CONCLUSION

The oxidation kinetics of D-galactose by IFC oxidant in a 50% (v/v) aqueous acetic acid medium were investigated at 303 K using spectrophotometric method. The reaction depicted a first-order relationship regarding [substrate], and [oxidant]. The presence of H^+ ions acted as a catalyst, also exhibiting a unit-order relationship with [H^+]. The oxidation process showed sluggishness in adding Mn^{2+} ions to the reaction blend, pointing towards the involvement of Cr(IV) species as an intermediate. The possibility of a free radical intermediate was eliminated, as there was no observed polymerization of acrylonitrile. Considering all the above facts, it was proposed that protonated IFC would interact with D-galactose and form an ester-like intermediate, which further undergoes a hydride ion elimination during the slowest step of the reaction to produce the final products.

5. CONFLICT OF INTEREST

Authors have no conflict of interest to declare.

6. ACKNOWLDGEMENT

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