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Comparative kinetics studies of ribose and galactose as eco-friendly reductants for leuco dye formation

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Abstract

Reduction of the dye is a significant process in the textile industry as it renovates the dye into water-soluble leuco dye for fixing it into a fibre. The current investigation explores the reduction kinetics of thionine dye (Th) in relation to the sugars consistent with the carbon chain to get a leuco dye state by an ecologically friendly reductant. For this purpose, galactose and ribose, the carbohydrate family with five and six carbon atoms, were selected, respectively. Reduction kinetics of Th with ribose and D-galactose monitored spectrophotometrically at λ_{max} 599 nm to optimize the different parameters as an essential step in leuco dye formation for dye fabrication. The results initially showed slow reduction kinetics, which later proceeded fast under different studied parameters, including the concentration of the dye, reductant, alkali and at different temperatures. The reaction followed pseudo-first-order kinetics in the presence of atmospheric oxygen. The results showed that the rate of the kinetics of Thionine with ribose was more influential than galactose keeping all parameters constant. A mechanism consistent with the above findings has been discussed in the relevant section of the paper.

Keywords: Thionine, Galactose, Ribose, Reduction, a shift in wavelength (λ).

1. INTRODUCTION

Thionine is a family member of thiazine dyes that have been extensively used in various fields of biochemical stainings due to its remarkable electrochemical properties[1–4**]**. It is also a redox indicator and reduced to leuco state or colourless Thionine. The reduction kinetics of Thionine is an important aspect of its behaviour in various applications[5,6]. The reduction of Thionine involves the transfer of electrons from a reducing agent to the thionine molecule, leading to a decrease in the oxidation state of the molecule[7-9]. The reduction kinetics of the dye is important in dye fabrication, where redox reaction is optimized for their application. It is optimized by various factors, including the reducing agent's nature and concentration, the solution's pH, and the temperature[10-13]. Several reports established the reduction kinetics of Thionine spectrophotometrically[14,15]. Moreover, several studies have also been employed to monitor the kinetics under various techniques, such as cyclic voltammetry[16,17], UV-visible spectroscopy[7,18], and stopped-flow techniques [19,20]. Understanding the reduction kinetics of Thionine is crucial for developing various electrochemical and analytical techniques. For instance, Thionine has been used as a mediator in enzyme-based biosensors to enhance the electron transfer rate between the enzyme and the electrode[21,22]. Additionally, the reduction kinetics of Thionine can be used to study the electron transfer reactions that occur in biological systems, such as photosynthesis and respiration[23**]**.

In this context, a comprehensive study of the reduction kinetics of Thionine can provide valuable insights into the underlying mechanisms of electron transfer reactions. This knowledge can lead to using the dyes in a more eco-friendly way for textile dye fabrication, where eco-friendly reducing agents are used to convert the dye molecule into colour less in the first step of the dye that diffuses easily with the fibre in the presence of atmospheric oxygen and fixed the colour on it the second step [20,23**]**. The redox reaction of thiazine dyes frequently occurs on a time scale of a few seconds to minutes[7,24]. Thionine dye was selected as the simplest dye molecule in dyes and pigments. Thionine exists as monovalent cations in the ordinary pH region, and their redox reactions are reversible. Since leuco Thionine (LTh) are very unstable and easily oxidized by coexisting oxygen in solution, few studies on the reaction of these compound with electron acceptors have been previously reported [25,26**]**. Because of the biological importance of carbohydrates and dyes, a detailed literature survey revealed that the kinetics of oxidation of carbohydrates; and kinetics of reduction of dyes had been studied widely, using various organic and inorganic compounds[27,28]. The current investigation provides the replacement of sodium sulfide for dyeing process with pentoses and hexoses in the alkaline medium. It was observed that sugars are effective in reducing the dye and dye becomes more soluble in water for dyeing process[29]

Therefore, this study aims to explore Thionine's reduction kinetics in the presence of two different reducing agents, galactose and ribose. The reduction kinetics was monitored under different parameters, including the concentration of dye and reductants, pH and temperature, followed by quantitative interpretation and spectrophotometrically for quantitative

determination. Results were discussed in relation to the effects of the concentration of galactose, Th, NaOH, temperature, and ionic strength of the medium. Activation parameters have been computed, and a mechanism has been proposed based on the above investigations.

2. MATERIAL AND METHODS

All reagents used in the experiment were purchased from Sigma and Aldrich and used without further purification. The experimental procedure involved the preparation of an aqueous methanolic solution containing D-galactose, Ribose, sodium hydroxide, and Thionine (E-Merck) at known concentrations. The solution was made in a 1:1 molar ratio of water to methanol, and the ionic strength was maintained using potassium nitrate. Kinetics measurements were performed by varying one species in the reaction mixture while keeping the other two species constant at given concentrations. Three sets of reaction mixtures were prepared and mixed. The progress of the reaction was monitored by recording the change in optical density at the maximum absorption wavelength (λmax=599 nm) using a UV-visible spectrophotometer (Shimadzu 1601).
The reaction order under different operational and activation parameters was determined by measuring the specific reaction rate at various temperatures and different ionic strengths. The percentage decrease in absorbance was calculated using the formula % of the decrease in absorption = $[(A_f - A_i)/A_f] \times 100$, where A_i and A_f represent the initial and final absorption, respectively. The UV/Visible spectrophotometer (Shimadzu 1601) was used throughout the experiments to measure the changes in absorption.

3. RESULTS AND DISCUSSION

The kinetics of the reduction of Thionine with D-galactose and Ribose was investigated by monitoring the change in optical density at λ max 599nm at different operational parameters. The investigation results were reported in Table 1, which indicated that the reaction followed pseudo-first-order kinetics for the dye, reductants, and alkali [30]. The redox reaction of the dye was investigated under aerobic conditions, and it was observed that initially, the reduction proceeded very slowly, as shown in Figure 1. However, there was a sharp decrease in optical density after some time, followed by a slow reduction process. Additionally, it was noted that the absorption value of the dye decreased with irradiation time, indicating that the dye converting into leuco dye. The results showed that both sugars acts as a best reducing agent for mainting the leuco state of the dye in presence of oxygen throughout experiment with appropriate amount. This observation may have important implications for reducing sugars to use in different vat dyes as ecofreindly reductants.

Table 1 First Order kinetics of Thionine at a different operational parametersTemperature = 30°C

$[Th^+]\ 10^5$ mol.dm^{-3}	**[NaOH] mol.dm^{-3}**	**[conc. Of reductant] 10^2 mol.dm^{-3}**	**Ribose $[dx/dt]10^4$ mol.dm^{-3}.cm^{-3}**	**Galactose $[dx/dt]10^4$ mol.dm^{-3}.cm^{-3}**	**Ribose 10^2k s^{-1}**	**Galactose 10^2k s^{-1}**
5.53	1.27	1.71	16	14	7.7	4.9
5.53	1.27	1.57	12	12	8.1	4.7
5.53	1.27	1.49	11	10	9.2	3.7
5.53	1.27	1.35	10	9	4.2	4.0
5.53	1.27	1.28	8	8	5	2.7
5.53	1.46	1.42	10	9	5.3	5.3
5.53	1.39	1.42	8	8	5.2	5.1
5.53	1.33	1.42	6	6	3.9	3.9
5.53	1.2	1.42	6	6	4.2	1.8
5.53	1.14	1.42	3	4	1.18	1.7
7.91	1.27	1.42	8	7	3.5	3.2

7.12	1.27	1.42	7	6	2.7	1.9
6.33	1.27	1.42	7	6	4.4	2.7
5.53	1.27	1.42	5	5	4.2	2.3
4.74	1.27	1.42	5	4	4.2	4.2

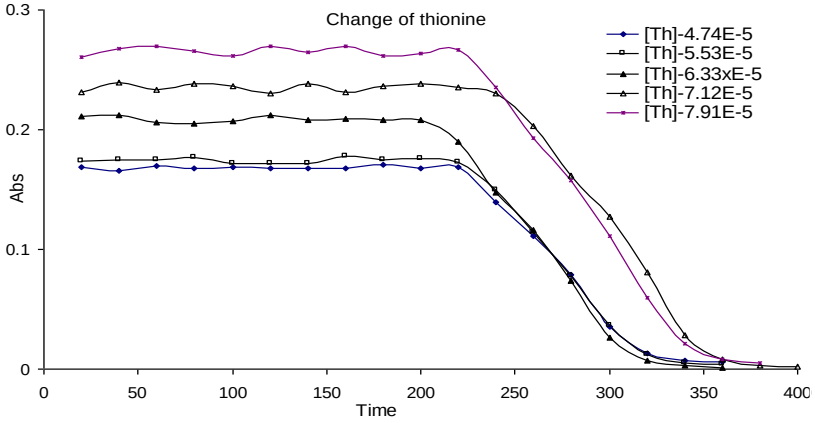

Figure 1: A plot of Absorbance Vs time (S) of reduction of Th with different concentrations of [Thionine]= $4.74x10^{-5}$-$7.91x10^{-5}$ $mol.dm^{-3}$, [NaOH] = 1.27 $mol.dm^{-3}$, [Ribose] = 1.43 $mol.dm^{-3}$ and Temperature = 303 K

3.1 Effect of dye, reductant and pH of the medium

The change in the absorption values was monitored at regular intervals, and a representative plot of optical density versus time was shown in Figure 1. Results reported in the Table 1, showed that the Thionine's reduction rate increased with an increase in the concentration of Thionine while keeping other parameters constant Figure-2. The decrease in the rate constant (k) with an increase in dye concentration showed an inverse relationship between the two. This suggested that fewer protons available for absorption to dye molecules inhibited the reduction rate. It was also observed that at the maximum concentration of reductant, there was a maximum percentage decrease in absorption, as shown in Table 2 and Figure 3, which was in accordance with the earlier report [30]that an increase in the concentration of reductant used caused an increase in the redox potential of the reducing systems[30]. In contrast, at high concentrations of Thionine, there was very less absorption of radiation, as shown in Table 3. It is also possible that at high concentrations of Thinone, the reaction becomes limited by other factors, such as mass transfer or the availability of reactive sites on the dye molecules. The study also showed that the rate of decoloration was directly proportional to the concentration of reductant (Ribose and Galactose). Overall, these studies demonstrate that the decoloration of thionine dye by galactose and ribose proceeds through a pseudo first-order kinetic mechanism and is influenced by the concentrations of the dye, reductant, and alkali. The reaction is thermally activated with a relatively low activation energy, and the rate of decoloration can be optimized by controlling the concentration of the reductant and temperature.

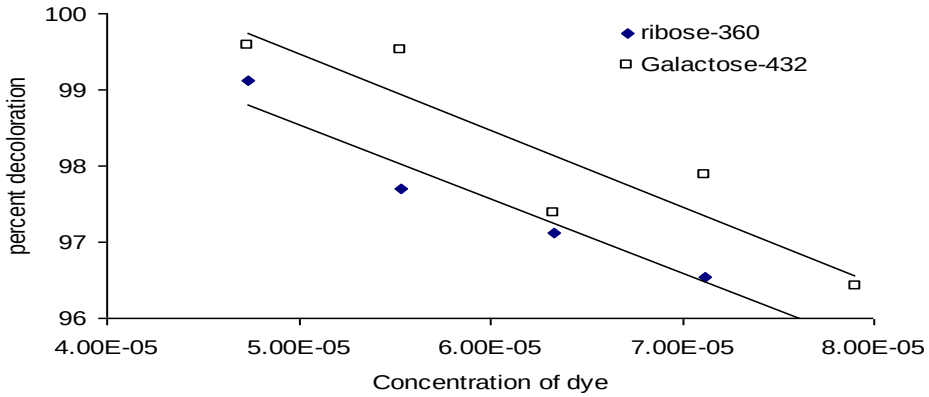

Figure 2: Effect of dye concentration on photo decoloration of Thionine with galactose and ribose.

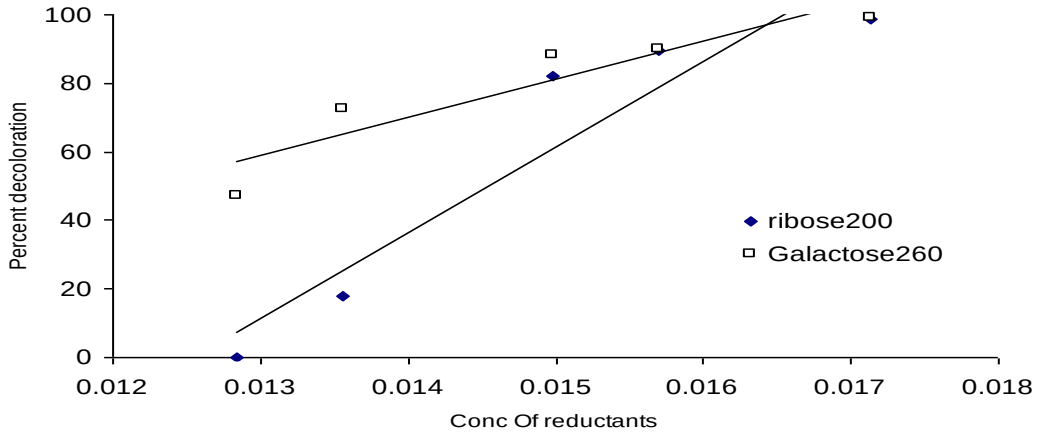

Figure 3: Effect of concentration of reductants on photo decoloration of Thionine with galactose and ribose.

Table 2 Dcoloration of Thionine at fixed time at different concentration of reductant

[Galactose] & [Ribose] 10^2 mol.dm^{-3}	**% decoloration at fixed time**	
	Ribose	Galactose
1.71	98.81	97.55
1.57	89.59	89.00
1.49	82.25	71.58
1.35	57.89	59.70
1.28	48.21	44.44

Table 3 Decoloration of Thionine at fixed time at different concentration of Thionine

[Thionine] 10^5 mol.dm^{-3}	**% decoloration at fixed time**	
	Ribose	Galactose
7.91	95.83	95.37
7.12	96.53	96.58
6.33	97.92	97.52
5.53	98.70	98.04
4.74	99.12	98.42

Table 5 illustrates how small amounts of base had an impact on pH values and revealed that the reduction was more pronounced in a higher pH range as shown in Figure-3. where it is visible that the reduction occurs more quickly and that the addition of a tiny amount of base causes the equilibrium to shift to the left. The regeneration of the oxidised form of Thionine was detected using the following process. In basic conditions, Thionine accepts an electron from the reductant and forms the thionine radical anion. The thionine radical anion reacts with another molecule of Thionine to form leucothionine. The leucothionine further accepts an electron and forms dihydroleucothionine, which is also colorless. The dihydroleucothionine reacts with oxygen in the air and undergoes auto-oxidation to form Thionine again. The addition of acid shifts the equilibrium towards the left-hand side, leading to the regeneration of Thionine from the reduced form.

Overall reaction:

Thionine + Reductant $+OH^{-1}\longrightarrow$ Leucothionine $\longrightarrow$ Dihydroleucothionine $\longrightarrow$ Thionine + O_2

3.2 Effect of ionic strength

The reaction rate between two uncharged molecules or between an ion and a molecule is typically only slightly affected by adding salt. The ionic strength ranged varied from 0.05 to 0.2 mol•dm^{-3}. The ionic strength of the medium experiments was conducted by varying the changing initial KNO_3 concentrations, constant thionine and reductant concentrations as shown in Table- 6, and various temperatures. The decoloration rate rises over time with different KNO_3 concentrations because the reaction rate between charged species is affected by ionic strength, which may be attributed to the dye reduction being more favourable in the presence of NO^{3-}. Table-6 demonstrated that at temperatures between 20 and 40 °C and varied ionic strengths, the apparent rate constant of dye decoloration rose. However, the apparent rate constant decreased at higher temperatures and stronger ions. Additionally, the reduction was more prominent in a higher pH and found positive ionic strength.

Table 4 Thermodynamic activation parameters of reduction of Thionine with galactose and ribose

Galactose	E_a 71.15kJ/mol									
Ribose	E_a 57.87kJ/mol									
	Temperature									
	303 K		**308 K**		**313K**		**318 K**		**323 K**	
	Gal	**Rib**	**Gal**	**Rib**	**Gal**	**Rib**	**Gal**	**Rib**	**Gal**	**Rib**
Δ H kJ/mol	68.63	55.36	68.59	55.32	68.55	55.26	68.51	55.23	68.47	55.19
- Δ S J/mol.K	57.74	99.42	57.87	99.55	58.00	99.69	58.14	99.82	58.27	99.95
Δ G kJ/mol	86.13	85.48	86.42	85.98	86.71	86.48	87.00	86.98	87.29	87.48

Table 5 Decoloration of Thionine at fixed time at different concentration of NaOH

[NaOH] mol.dm^{-3}	**% decoloration at fixed time**	
	Ribose	Galactose
1.46	97.6	96.99
1.39	97.58	97.02
1.33	88.12	87.74
1.20	86.54	81.22
1.14	52.01	47.77

Table 6 Effect of temperature on dye reduction at different ionic strength. [Thionine] = 6.33x10^{-5} mol dm^{-3} [Galactose] = 1.42x10^{-2} , [Ribose] = 1.42x10^{-2} mol dm^{-3}mol dm^{-3} [NaOH] = 1.27mol dm^{-3}

Ionic strength	**Temperature**									
$\sqrt{\mu}$	**303 K**		**308 K**		**313K**		**318 K**		**323 K**	
10^{2}mol.dm^{-3}	10^2k s^{-1}		10^2k s^{-1}		10^2k s^{-1}		10^2k s^{-1}		10^2k s^{-1}	
	Gal	Rib	Gal	Rib	Gal	Rib	Gal	Rib	Gal	Rib
5.00	0.82	1.49	1.94	1.86	3.77	3.3	2.81	2.55	2.49	2.74
8.75	0.93	1.33	1.87	2.03	3.67	4.95	3.18	4.72	3.16	2.5
12.50	0.99	1.7	1.89	2.56	3.93	3.1	3.22	4.76	3.09	1.73
16.25	1.13	1.59	2.29	3.38	4.21	3.83	3.15	3.11	2.66	3.24
20.00	1.15	1.48	2.29	4.12	3.60	4.31	3.53	3.91	3.03	3.98

3.3 Effect of temperature

The reduction kinetics investigated at several temperature and results are reported in the Table-4 while reperesentative plots are shown in Figure 4. It was observed that reduction was depended upon temperature as reported ealier [30] indicating the reaction was thermally activated. The activation energy (E_a) was determined from the rate constant (k) temperature dependence using the Arrhenius equation[7]. The activation energy was found 71.15 kJ/mol and 57.87 kJ/mol for ribose and galactose, respectively (Table 4,5), indicating that the reaction has relatively low activation energy and is likely to proceed at a reasonable rate at ambient temperatures[5]. The Ea shows that the activation energy of ribose is less than that of galactose, supporting that the scarbon chain effectively involve in the reduction of the dye. The entropy of activation (ΔS) for the reduction of Thionine with D-galactose and ribose is -57.74 $J•K\text{-}1•mol^{-1}$ and 99.42 $J•K\text{-}1•mol^{-1}$ at 303K for galactose and ribose, respectively. These values indicate that the activated complex was in a highly solvated form. If ΔS is negative, either the solvent molecules are tightly bound to the OH^- bond, the site of oxidation, or there is a reduction in the degree of freedom due to the creation of a rigid activation complex that causes substantial reorientation of the solvent molecules. The transition state is substantially saturated, as evidenced by the relatively high values of enthalpy or activation ΔH, free energy of activation ΔG, and energy of activation (E_a). Additionally, it demonstrates that, compared to the reactants, the rate-determining state is less disordered-orientated. The positive value of ΔH 68.63 $kJ•mol^{-1}$ and 55.36 $kJ•mol^{-1}$ demonstrated that entropy, rather than enthalpy, was the drive behind the synthesis of the complex that includes one charged species and solvent molecules for dye reduction. The activation energy was found to be significant, suggesting that the reaction rate was highly dependent on temperature. The entropy change was negative, indicating that the reaction was not spontaneous. Therefore, an increase in the temperature of the reaction was required to overcome the activation energy barrier and initiate the reaction.

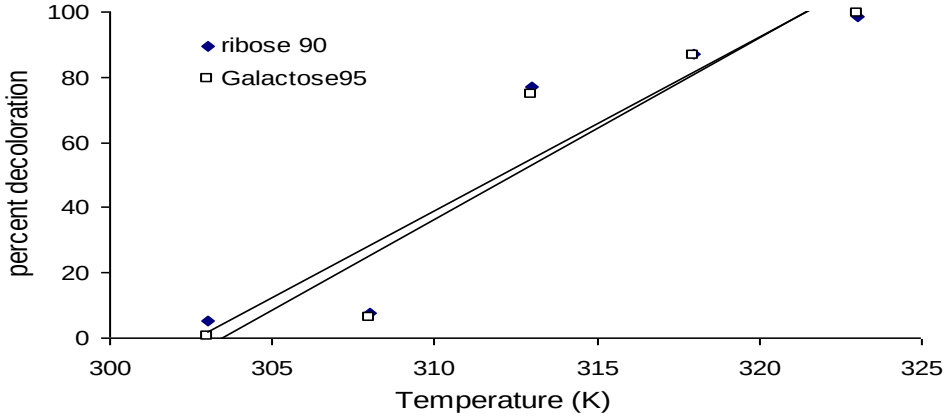

Figure 4: Effect of temperature on photo decoloration of Thionine with galactose and ribose.

4. CONCLUSION

The study was conducted for the first time with Thionine and sugars as reducing agents related to the carbon chain to optimize the reduction kinetics parameters like concentration of dye, reductant, pH, ionic strength and temperature. It was concluded that both sugars might act as the best eco-friendly reducing agent as they permit the stable state of the leuco-soluble form of the dye in the presence of atmospheric oxygen with an appropriate amount of reductants, pH and dye concentration. The leuco dye formation is the first step in dye processing with sugars, showing that reducing sugars can take the place of other environmentally toxic reductants like sulphide reductants commonly used in first step of dye fabrication. However, more work is required to validate the further second step of the oxidized form of the dye required for fixing in the fabrics

In conclusion, this study provides insight into the mechanism of thionine reduction with ribose, galactose and the factors that affect the reaction rate. These findings have potential applications in ribose and further show that ribose is a better-reducing agent than galactose.

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