

Reduction of Methylene Green by EDTA: A relation between dielectric constant of medium and activated state

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Abstract: Kinetics of an alkaline reduction of the methylene green with ethylenediaminetetraacetic acid (EDTA) as a role of dielectric constant of the medium with anecdotal ionic strength in a diverse solvent system (aqueous mixtures of methanol) (10-30%) was studied by measuring the specific rate constant of the reaction spectrophotometrically at $\lambda_{\text{max}} = 660\text{nm}$. An effort has been made to give an elucidation of vital role of dielectric constant of the medium captivating into reflection of single sphere and double sphere complex in reaction assortment. This investigation leads to disclose that single sphere complex of the dye and reductant was found to be the most suitable complex existed in a varied organic solvent. The deviation of the theoretical values from experimental results for single sphere and double sphere complex model in the presence of an alkali and nitrate ions were justified through HPLC analysis. HPLC analysis recommended that a considerable amount of the dye degrades in the existence of nitrate ion and alkali and additional peaks which may be of by-product were obtained. This leads to confirm the non identical values of single sphere and double sphere model in the occurrence of nitrate and an alkali. Rate of deletion of color showed a linear liaison with respect to water content below 30% and temperature between 20-40°C whereas an increase in the concentration of organic solvent showed the inhibition of dye decoloration at given optimum condition. Therefore study was restricted up to 30% of methanol binary mixtures. A mechanism of reduction of dye has been proposed based on verdict.

Keywords: Methylene green, EDTA, solvent, dielectric constant, single sphere complex, double sphere complex, HPLC, degrade, decoloration.

INTRODUCTION

The power of *solvent* dielectric relaxation on the velocity of an electron transfer (ET) at an electrochemical interface was addressed through both experiment and model calculations in water-ethylene glycol (EG) (Pavel *et al.* (2010). Three groups of experimental results were determined to characterize the mixed-solvent system (dielectric spectra in the frequency range 0.1-89 GHz), the mercury/solvent interface (electro capillary data), and the ET kinetics (dc polarography of peroxodisulphate reduction). The results demonstrate that the observed anomalous “viscosity effect” may be explained qualitatively by an increased contribution of the fast relaxation mode at high EG contents. Saeed *et al.* (2010) reported a bathochromic shift with the decrease in polarity of solvent in different percentages of methanol (0-50%). It was also observed that an increase in the rate of reaction was the results of an increase in an alcoholic content which was affected by potassium iodide salt. Raiman *et al.* (2001) observed the influence of pH and dielectric constant on spectral behavior of pinacyanol using spectrophotometer. Clodoaldo and Vanderlei (2001) investigated the effect of solvent polarity via solvate chromo dyes spectrophotometrically and premeditated the transition energies of dyes in each solvent. The attraction of columbic energy of activation for a reaction can be calculated from the equation with a change in the

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dielectric constant of the solvent. Uddin and Kazmi (2003) reported the reaction involving potassium peroxydisulfate and potassium iodide in a sequence of water-acetone mixtures. Results showed that the explanation of the activated complex could best be given by the single sphere model. Leen *et al.* (2010) investigated the effect of the molecular structure on the visible absorption and fluorescence emission properties of BODIPY as a role of solvent through new, generalized treatment of the solvent effect. Solvent polarizability was the key factor responsible for the small solvent-dependent shifts of the visible absorption and fluorescence emission bands of the dyes. The configuration and solvent effect on the photo stability of a chain of indolenine cyanine dyes have been investigated by Chen *et al.* (1999). It was found that the photo stability of the dyes did not increase with an increase of the central conjugated chain length and/or substituent groups on the heterocyclic ring using UV spectral data. Results showed that the photo stability of indolenine cyanine dyes in solution was directly related with the $E_T(30)$ values of solvents.

The aims and objectives of present work were to investigate the effects of diverse solvent system on rate constant of methylene green and EDTA in existence of salt and alkaline medium. An endeavor has been made to give an explanation of crucial role of dielectric constant of the medium taking into consideration of single sphere and double sphere complex in reaction mixture and a

possible mechanism of dye reduction with EDTA, based on above observation.

MATERIALS AND METHODS

All reagents used were Analar grade, purchased from E-Merck and used without further purification. Stock solution of MG concentration 1×10^{-4} mol.L⁻¹ was prepared in 100 ml of deionized water. Stock solutions of EDTA of 0.1 mol.L⁻¹, NaOH of 0.5 mol.L⁻¹ and KNO₃ of 1.0 mol.L⁻¹ were also prepared in deionized water (Azmat *et al.*, 2006). The experimentation was planned for a 3 hr laboratory sitting. Mandatory amount of dilutions were made at the time of experiments. Measurements of reaction kinetics include various factors like concentration of dye, EDTA, NaOH, KNO₃ and effect of solvent (10-30% methanol) on the activated state through spectrophotometer. Data interpretation was done by linear regression method.

Kinetics measurements

Sequencings of water – methanol mixtures for reaction between methylene green and EDTA were carried out at a wavelength of 660nm. The ionic strength of reaction mixture was altered by varying KNO₃ concentrations. Rate constants at zero ionic strength were calculated (Azmat *et al.*, 2006). The values of dielectric constant of methanol water mixtures were (w/w) tabulated in the table 1.

Table 1: Dielectric constant (ϵ) values of methanol water mixture at various temperatures

Given by Akerlof (1932)

Temperatures °C	Dielectric constant (ϵ) 10% methanol	Dielectric constant (ϵ) 20% methanol	Dielectric constant (ϵ) 30% methanol
20°C	75.84	71.02	66.01
25°C	74.045	69.17	64.29
30°C	72.37	67.48	62.71
35°C	70.5329	65.80	61.19
40°C	68.90	64.13	59.53

Table 6: A Comparison of “k” rate constant with Literature

Comparison of dyes		Literature	Comparison of reductant			Literature
$k \times 10^3$ (s ⁻¹) MG (current work)	$k \times 10^3$ (s ⁻¹) MB (literature value)		$k \times 10^3$ (s ⁻¹) SO ₃ ⁻² (literature value)	$k \times 10^3$ (s ⁻¹) glucose (literature value)	$k \times 10^3$ (s ⁻¹) EDTA (current work)	
9.0	56	Snehalatha <i>et al</i> (1997)	0.61	3.3	4.0	Jonnalagadda, <i>et al</i> (2000) Azmat <i>et al</i> (2006)
9.0	62		1.30	3.4	6.0	
9.0	59		2.27	3.9	9.0	
8.5	64		3.10	4.2	10.0	
8.7	64		3.95	4.4	11.0	

HPLC and Mass spectrometric analysis

HPLC analysis of reaction mixture was conceded out for the divergence of experimental values from theoretical ones, of activated complex existing in the mixed solvent system. For this purpose 50 ml of dye solution were taken either at time zero (right after addition of EDTA and no UV irradiation) or at 30 min of UV irradiation after addition of EDTA, and the organic component of the mixture was extracted using chloroform. The organic layer (chloroform) was extracted again for a second time with chloroform, and then dried in a rotary evaporator. The dried substance (dye and/or degraded by-products) was then dissolved in 100% methanol. The reaction mixture of MG was injected into the column which appears at 2.86 min retention time. The samples were run on C⁸-Eclipse column (Agilent) using a 0-100% methanol (water as the base mobile phase) gradient over 30 min, at 1 ml/min, and analyzed by measuring the absorbance at 280 nm. Isolated products were subjected to mass spectrometry by the method described by Azmat (2009).

RESULTS

Results of the kinetics measurements for the reduction of methylene green by EDTA in 10 to 30% methanol water system at 303 K are being reported in the tables 2-5. Rate constants for this reaction was calculated by plotting $\ln A$ vs time for varying concentrations of reactants viz. MG, EDTA, NaOH and KNO₃ at different composition of solvent i.e. methanol water mixtures from the slopes of the plots. It was noted that rate constants at a constant ionic strength increases with an increase in percentages of the organic solvents from 10 to 30%. It was also noted that for the identical value of dielectric constant of the medium, an increase in the concentration of EDTA, NaOH, MG, and KNO₃ increase the rate constant in case of both reductant and base but almost independent upon the concentration of dye. The values of k_0 were determined through Debye Huckle Bronsted equation, which was a connection between ionic strength and rate constant (equation 1). Effect of dielectric constant on the rate constant was investigated in the existence of methanol-water mixture at temperature 303 K. The relationship between ionic strength and the rate

constants is given by the Debye Huckle Bronsted (Debye 1923) equation (1)

$$\ln k = \ln k_0 + \frac{e^3 \left(\frac{8\pi N}{1000} \right)^{\frac{1}{2}}}{(\epsilon K T)^{\frac{3}{2}}} \cdot Z_A Z_B (\mu)^{\frac{1}{2}} \quad (1)$$

where “k” is the rate constant at zero ionic strength, e, N, ϵ , k, T are electric charge, Avogadro’s Number, dielectric constant of the medium, Boltzmann’s constant and absolute temperature respectively. Z_A and Z_B are the charges of A and B respectively. The values of k (s^{-1}) were calculated using equation (1) and tabulated in tables 2-5. The plots of $\log k_0$ against the reciprocal of dielectric constant are straight line with positive slopes. Representative plots are shown in the figs. 1-4 respectively. The experimental values of $r^\#$ and r_{AB} are calculated from the slopes of straight lines using equations 2 and 3 are tabulated in table 7. Theoretical results were justified through HPLC and mass spectrometry and proved complex kinetics involved in the degradation of dye EDTA complex.

Table 2: Rate measurements data for MG methanol water mixture at 30°C at different ionic strength

ϵ	$k \times 10^{-3}$ (s^{-1})	k_0 (s^{-1})
$\sqrt{\mu}_1 = 1.3039210 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	3.9	3.3307
67.48	4.1	7.3905
62.71	4.3	18.524
$\sqrt{\mu}_2 = 1.3039248 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	3.5	2.9876
67.48	3.7	6.6657
62.71	4.6	19.805
$\sqrt{\mu}_3 = 1.3039286 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	3.3	2.8169
67.48	3.8	6.8461
62.71	4.5	19.375
$\sqrt{\mu}_4 = 1.3039325 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.5	2.1352
67.48	3.4	6.1291
62.71	4.8	20.680
$\sqrt{\mu}_5 = 1.3039363 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.0	1.7073
67.48	2.9	5.2248
62.71	3.3	14.2091

$C_{MG} = (1 \times 10^{-5} - 5 \times 10^{-5}) \text{ mol.L}^{-1}$, $C_{EDTA} = 1 \times 10^{-4} \text{ mol.L}^{-1}$, $C_{NaOH} = 1.5 \text{ mol.L}^{-1}$, $C_{KNO_3} = 0.2 \text{ mol.L}^{-1}$, Temperature = 303 K, Solvent = (10% - 30%) Methanol.

Table 3: Rate measurements data for EDTA methanol water mixture at 30°C

ϵ	$k \times 10^{-3}$ (s^{-1})	k_0 (s^{-1})
$\sqrt{\mu}_1 = 1.30392 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.5	2.1350
67.48	4.8	8.6522
62.71	2.9	12.4930
$\sqrt{\mu}_2 = 1.30399 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.8	2.3921
67.48	2.9	5.2290
62.71	3.0	12.930
$\sqrt{\mu}_3 = 1.30407 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	3.1	2.6359
67.48	3.6	6.4577
62.71	3.8	16.2820
$\sqrt{\mu}_4 = 1.30415 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	4.2	3.5276
67.48	4.4	7.8962
62.71	4.6	19.7190
$\sqrt{\mu}_5 = 1.30422 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	4.6	3.9144
67.48	5.7	10.2330
62.71	6.4	27.4400

$C_{MG} = 1 \times 10^{-5} \text{ mol.L}^{-1}$, $C_{EDTA} = (1 \times 10^{-4} - 5 \times 10^{-4}) \text{ mol.L}^{-1}$, $C_{NaOH} = 1.5 \text{ mol.L}^{-1}$, $C_{KNO_3} = 0.2 \text{ mol.L}^{-1}$, Temperature = 303 K, Solvent = (10% - 30%) Methanol.

Table 4: Rate measurements data for NaOH methanol water mixture at 30°C

ϵ	$k \times 10^{-3}$ (s^{-1})	k_0 (s^{-1})
$\sqrt{\mu}_1 = 0.7072 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.0	0.077
67.48	2.0	0.116
62.71	2.8	0.261
$\sqrt{\mu}_2 = 0.8945 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.6	0.266
67.48	3.1	0.530
62.71	3.4	1.058
$\sqrt{\mu}_3 = 1.0489 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	3.5	0.798
67.48	5.0	2.080
62.71	3.4	2.850
$\sqrt{\mu}_4 = 1.1833 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	5.9	2.698
67.48	6.3	5.675
62.71	6.8	13.508
$\sqrt{\mu}_5 = 1.3039 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	6.6	5.363
67.48	6.9	12.430
62.71	9.2	39.630

$C_{MG} = 1 \times 10^{-5} \text{ mol.L}^{-1}$, $C_{EDTA} = 1 \times 10^{-4} \text{ mol.L}^{-1}$, $C_{NaOH} = (0.3, 0.6, 0.9, 1.2, 1.5) \text{ mol.L}^{-1}$, $C_{KNO_3} = 0.2 \text{ mol.L}^{-1}$, Temperature = 303 K, Solvent = (10% - 30%) Methanol.

Table 5: Rate measurements data for KNO₃ methanol water mixture at 30°C

ϵ	$k \times 10^{-3}$ (s ⁻¹)	k_0 (s ⁻¹)
$\sqrt{\mu}_1 = 1.3039 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	3.1	2.4672
67.48	5.2	9.3722
62.71	11.0	47.383
$\sqrt{\mu}_2 = 1.3784 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	3.0	3.767
67.48	4.4	12.1707
62.71	2.8	19.455
$\sqrt{\mu}_3 = 1.4492 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.6	4.7104
67.48	5.1	21.190
62.71	20.0	218.89
$\sqrt{\mu}_4 = 1.5166 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.6	6.677
67.48	4.6	28.16
62.71	9.3	156.87
$\sqrt{\mu}_5 = 1.5812 \text{ (mol dm}^{-3}\text{)}^{0.5}$		
72.37	2.5	8.97
67.48	3.7	32.844
62.71	4.5	114.90

$C_{MG} = 1 \times 10^{-5} \text{ mol.L}^{-1}$, $C_{EDTA} = 1 \times 10^{-4} \text{ mol.L}^{-1}$, $C_{NaOH} = 1.5 \text{ mol.L}^{-1}$, $C_{KNO_3} = (0.2, 0.4, 0.6, 0.8, 1.0) \text{ mol.L}^{-1}$, Temperature = 303 K, Solvent = (10% - 30%) Methanol

DISCUSSION

The effect of varying concentration of dye and reductant EDTA with constant concentration of other variable almost showed the same behavior as reported earlier in the literature (Saeed *et al.*, 2010 and Azmat *et al.*, 2008, Samanta *et al.*, 2011) for the similar value of dielectric constant of the medium i.e. an increase in the concentration of EDTA, increases the rate constant but it was self-governing for the concentration of dye (table 6). Rate of removal of color showed a linear relationship with respect to water content below 30% and temperature between 20- 40°C where as an increase in the concentration of organic solvent showed the inhibition of dye decoloration at given optimum condition. Therefore study was limited to 30% of methanol binary mixtures. A comparison of the rate constant “k” with earlier investigators reflects that the dye reduction linearly depend upon the concentration of EDTA as a reducing agent (table 6) in an aqueous mixed medium. Approximately similar reactions were observed in a mixed solvent system reflecting the dominant character of an aqueous medium.

Results tabulated in the tables 2-5 showed that the rate constant “k” at a constant ionic strength was inversely

related with the dielectric of the medium i.e. the rate of reaction between MG and the EDTA increases with an increase in the percentage of alcohol or decrease in the dielectric constant of medium. This inverse relation leads to conclude that there is an increase in the electrostatic interaction between the reacting ions at lower dielectric constant. That is, activity of the reactants increases with the decrease in the dielectric constant.

Table 7: Experimental values of r_{AB} (Å) and $r^\#$ (Å) at different $\sqrt{\mu}$ in Methanol–Water Media

$\sqrt{\mu} \times 10^{-4}$ (mol dm ⁻³) ^{0.5}	r_{AB} (Å)	$r^\#$
MG		
1.3039210	1.34	0.494
1.3039248	1.22	0.4295
1.3039286	1.20	0.4187
1.3039325	1.01	0.335
1.3039363	1.09	0.3690
Average	1.172	0.4092
EDTA		
1.30392	1.32	0.48
1.30399	1.37	0.50
1.304074	1.27	0.45
1.304151	1.35	0.49
1.304227	1.19	0.41
Average	1.30	0.46
NaOH		
0.7072	1.89	0.086
0.8945	1.68	0.069
1.0489	1.06	0.035
1.18300	1.43	0.054
1.3039	1.18	0.041
Average	1.44	0.057
KNO ₃		
1.3039	0.24	0.80
1.3784	0.53	1.42
1.4492	0.17	0.60
1.5166	0.28	1.52
1.5812	0.29	0.90
Average	0.30	1.04

The dependence of rate constant on dielectric constant was explained by the following Emeleus, & Anderson, (1961) and confirmed by Willson and Newall (1966)

$$\log k_o = \log k_\infty - \frac{e^2}{4.606 \epsilon KT} \left[\frac{(Z_A + Z_B)^2}{r^\#} - \frac{Z_A^2}{r_A} - \frac{Z_B^2}{r_B} \right] \quad (2)$$

$$\log k_o = \log k_\infty - \frac{Z_A Z_B e^2}{2.303 K T r_{AB} \epsilon} \quad (3)$$

where equations 2 and 3 are for single and double sphere complex model respectively. In the above equation k_o and k_∞ is the rate constants at zero ionic strength and infinite dielectric constant respectively. $Z_A = (+1)$ is the valence

of methylene green ion and $Z_B = (-2)$ is the valence of EDTA ion, $r^\#$ is the radius of the activated complex for single sphere model, r_A and r_B are the radii of the methylene green and EDTA ions respectively. r_{AB} is the distance between the centers of methylene green and EDTA ions in an activated complex for a double sphere model. Other terms have their usual meanings. The plots of logarithm of rate constant ($\log k_o$) against the reciprocal of dielectric constant are linear with positive slopes (figs. 1-4), which showed the rate constant increased with the decrease in the dielectric constant of the medium. The slopes indicate that the activated complex is more solvated than the reactants.

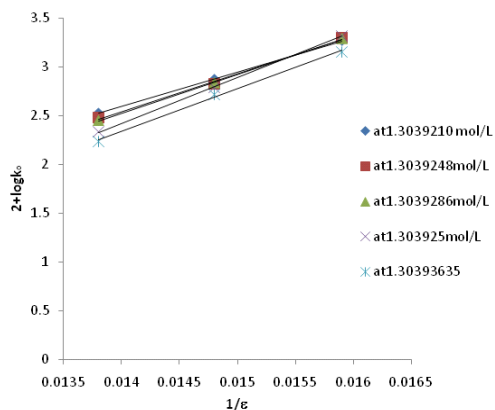


Fig. 1: Plot between $1/\epsilon$ vs. $2 + \log k_o$ for MG in methanol- water system at 30°C.

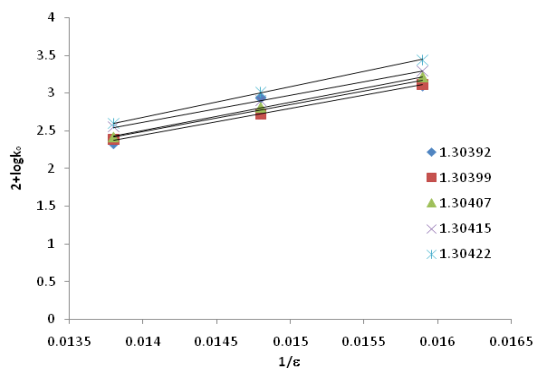


Fig. 2: Plot between $1/\epsilon$ vs. $2 + \log k_o$ for EDTA in methanol- water system at 30°C

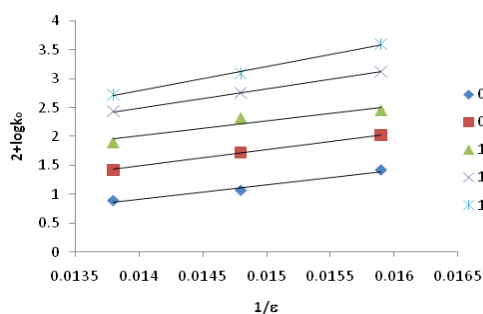


Fig. 3: Plot between $1/\epsilon$ vs. $2 + \log k_o$ for NaOH in methanol- water system at 30°C

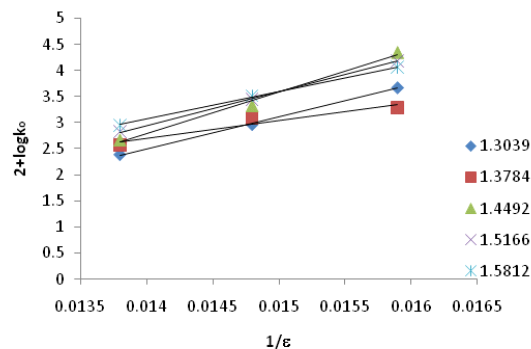


Fig. 4: Plot between $1/\epsilon$ vs. $2 + \log k_o$ for KNO_3 in methanol- water system at 30°C.

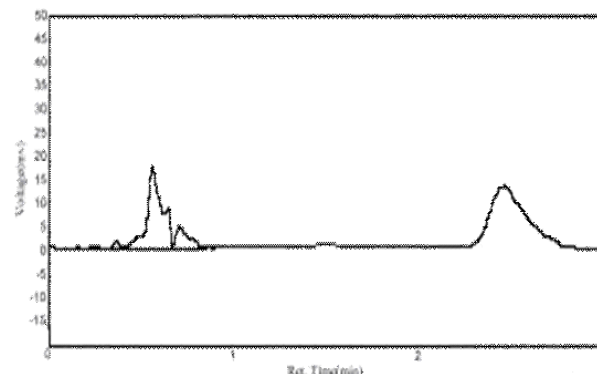


Fig. 5: HPLC Chromatogram for degradation of MG before irradiation in water.

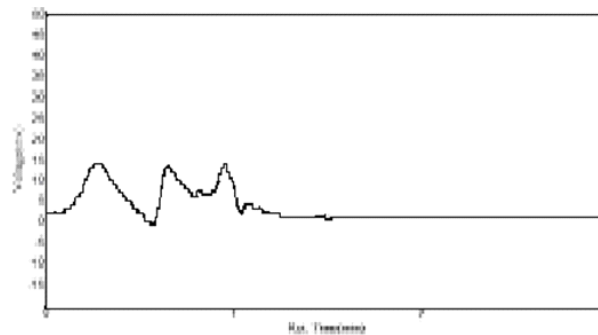


Fig. 6: HPLC Chromatogram for degradation of MG after irradiation of 30 min. in Water

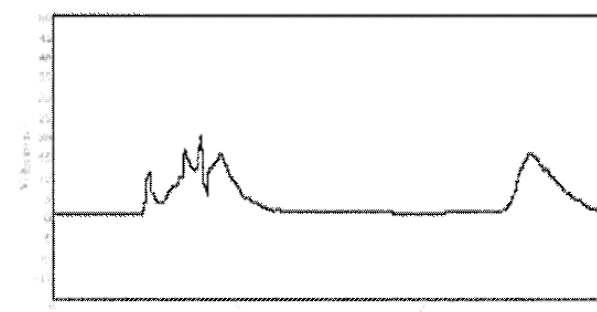


Fig. 7: HPLC Chromatogram for degradation of MG before irradiation in mixed solvent system.

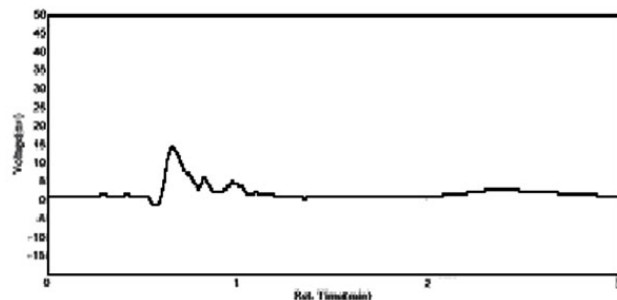


Fig. 8: HPLC Chromatogram for degradation of MG after irradiation of 30 min. in mixed solvent system.

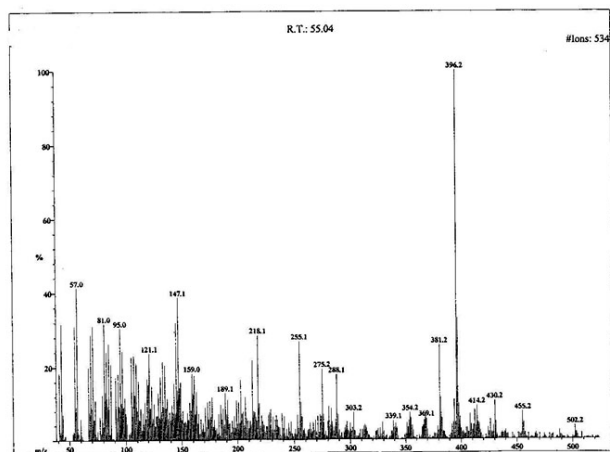


Fig. 9: Mass spectrograph of isolated product of reaction.

Radii of activated complex

The values of radii of activated complex calculated by using the equation (2 and 3) of single and double spheres model are tabulated in the table 7.

Table 8: Comparison of values of radii determined experimentally with theoretical values methanol water media

	Experimental				Theoretical
	MG	EDTA	NaOH	KNO ₃	
$r^\# (\text{\AA})$	0.409	0.46	0.057	1.04	6.8
$r_{AB} (\text{\AA})$	1.17	1.30	1.44	0.30	10.887

Theoretical calculation of radii of activated complex

The radius of methylene green ion was calculated based on the assumption that the volume of the molecule of a compound supposed to be spherical and is the sum of the volumes of individual's ions and determined as follows:

$$V_{C_{16}H_{17}N_4SO_2} = 16V_{C^{+4}} + 17V_{H^{+}} + 4V_{N^{+3}} + V_{S^{-2}} + 2V_{O^{-2}}$$

$$V = \frac{4}{3}\pi r^3$$

$$\frac{4}{3}\pi r^3 = 16 \times \frac{4}{3}\pi r^3 C^{+4} + 17 \times \frac{4}{3}\pi r^3 H^{+} + 4 \times \frac{4}{3}\pi r^3 N^{+3} + \frac{4}{3}\pi r^3 S^{-2} + 2 \times \frac{4}{3}\pi r^3 O^{-2}$$

$$r_{C_{16}H_{17}N_4SO_2}^3 = 16r_{C^{+4}}^3 + 17r_{H^{+}}^3 + 4r_{N^{+3}}^3 + r_{S^{-2}}^3 + 2r_{O^{-2}}^3$$

$$r_{C_{16}H_{17}N_4SO_2}^3 = 16(0.15)^3 + 17(2.08)^3 + 4(1.71)^3 + (1.84)^3 + 2(1.4)^3$$

$$r_{C_{16}H_{17}N_4SO_2}^3 = 0.054 + 152.98 + 20.00 + 6.22 + 5.488$$

$$r_{C_{16}H_{17}N_4SO_2}^3 = 184.743$$

$$r_{C_{16}H_{17}N_4SO_2} = 5.69538 \text{ \AA}$$

Similarly the value of radius of EDTA⁻² ion was calculated as 5.192 Å. The radius of activated complex for double sphere model (r_{AB}) is calculated based on the assumption that the radius of activated complex is the sum of radii of reactants.

$$r_{AB} = r_{MG} + r_{EDTA^{-2}}$$

$$r_{AB} = 5.69538 + 5.192$$

$$r_{AB} = 10.887 \text{ \AA}$$

The radius of activated complex for single sphere model ($r^\#$) is calculated on the basis of assumption that the volume of single sphere complex is the total of volumes of reactants therefore:

$$V^\# = V_A + V_B$$

$$\frac{4}{3}\pi (r^\#)^3 = \frac{4}{3}\pi (r_A)^3 + \frac{4}{3}\pi (r_B)^3$$

$$(r^\#)^3 = r_A^3 + r_B^3$$

$$(r^\#)^3 = (5.695)^3 + (5.192)^3$$

$$(r^\#)^3 = 352.234$$

$$r^\# = 6.8 \text{ \AA}$$

In all calculations, the values of the radius of the individual ions were taken from Emeleus (1961) and confirmed by Wilson (1966).

Comparison of the theoretical and experimental values of radii of activated complex

A comparison of the theoretical and experimental values of $r^\#$ and r_{AB} , for single and double sphere model shown in the table 8, suggested that the shape of the activated complex was more nearer to single sphere activated complex in the reaction at 303 K. This recommended that the description of the activated complex could best be explained by single sphere model for the reduction of MG⁺ and EDTA⁻² ions while the decrease in the values of $r^\#$ from the theoretical values were tried to explain through HPLC and mass spectrograph which may be attributed to the degradation of activated complex.

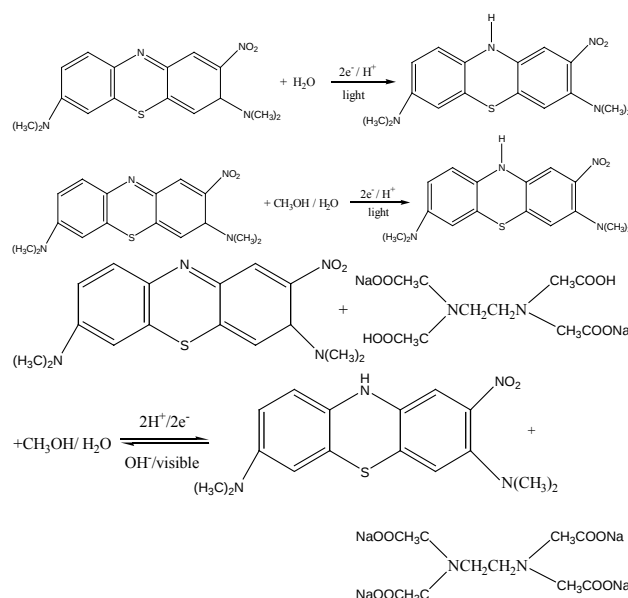
Verification of results through HPLC and Mass spectrometric analysis

In order to explain the non- identical values of ionic radii for single sphere and double sphere model to that of

theoretical ones, HPLC analysis was performed to check whether dye degraded into small fragments to form bi-products. During photolytic treatment, sample of dyes solution were taken at specific interval and subjected to HPLC analyses which were shown in the figs. 5-9. Figs. showed that the considerable amount of dye was degraded with regular time interval and additional peaks presumably bi-products were produced. So it may conclude that overall photolytic reduction of the dye may be due to the reaction of the dye with hydroxyl radical generated in an alkaline medium. So decoloration is most likely due to the actual dye degradation with reduction followed with reduction (Abdullah 2006). These peaks were also confirmed by further mass spectrometry which showed m/z at 381.2, 396.2, 414.2, 430.2, 455.2 and 502.2. These molecular ions peak suggest that single sphere activated complex degraded into smaller fragments during complex formation in the activated state that result in the decoloration of dye and non-identical experimental values to that of theoretical values.

Mechanism of dye reduction in solvent and EDTA

The reduction of MG with EDTA in mixed solvent system and EDTA may be explained as follows.



CONCLUSION

It was concluded that dielectric constant of the medium plays a imperative role in dye reduction which tends to decrease in mixed solvent system and single sphere activated complex more likely be existed in the transition state before leuco dye formation

REFERENCES

Abdullah FH, Rauf MA and Ashraf SS (2006). Kinetics and optimization of photolytic oxidation of coomassie brilliant blue with H_2O_2 . *Dyes and Pigments* **69**: 74-78.

- Akerlof GJ (1932). The dielectric constant of water at a high temperature *Am. Chem Soc.*, **54**: 4130.
- Azmat R (2009). Reduction of Methylene Blue with Reducing Sugars. Publisher: VDM Verlag Dr. Müller EK
- Azmat R, Ahmed S, Qureshi S, Mohammed FV and Uddin F (2006). Aerobic oxidation of D-glucose by methylene green in alkaline aqueous solution by visible spectrophotometry. *J. Appl. Sci.*, **6**: 2784-2788.
- Azmat R, Qamar N, Saeed A and Uddin F (2008). Reduction of methylene green by EDTA: Kinetic and thermodynamic aspects. *Chinese J. Chem.*, **26**, 631-634.
- Chen P, Sun S, Hu Y, Qian Z and Zheng D (1999). Structure and solvent effect on the photostability of indolenine cyanine dyes *Dyes and pigments* **41**: 227-231.
- Clodoaldo M and Vanderlei GM (2001). An easy and versatile experiment to demonstrate solvent polarity using solvatochromic dyes. *J. Chem. Ed.*, **78**(5): 649-651.
- Debye P and Huckel E (1923). The theory of electrolytes. *Physikalische Zeitschrift*, **24**(9): 185-206.
- Emeleus HJ and Anderson JS (1961). Modern aspects of Inorganic Chemistry, English Language Book Soc., London.
- Jonnalagadda SB and Gollapalli NR (2000). Kinetics of reduction of toluidine blue with sulfite-kinetic salt effect in elucidation of mechanism *J. Chem. Edu.*, **77**(4): 506-508.
- Laidler KJ (1965). Chemical Kinetics. 2nd Ed, McGraw Hill Book Company.
- LeenV, Qin W, Yang W, Jie C, Xu C, Tang X, WL, Robeyns R, Meervelt LV, Beljonne D, Lazzaroni R, Tonnele C, Boens N and Dehaen W (2010). Synthesis, spectroscopy, crystal structure determination, and quantum chemical calculations of BODIPY dyes with increasing conformational restriction and concomitant red-shifted visible absorption and fluorescence spectra. *Chemistry an Asian J.*, **5**(9): 2016-2026.
- Pavel A, Zagrebina RB, Renat, Nazmutdinov R and Galina A Tsirlina (2010). Dynamic solvent effects in electrochemical kinetics: Indications for a switch of the relevant solvent mode. *The J. Phy. Chem. B.*, **114**(1): 311-320.
- Raimon S, Liucia F and Joan E (2001). Influence of dielectric constant on the spectral behavior of pinacyanol. A spectrophotometric experiment for physical chemistry. *J. Chem. Ed.*, **78**(2): 243-244.
- Saeed R, Ashfaq M, Uddin F and Khan SA (2010). Effect of solvent on kinetics of reduction of methyl violet with potassium iodide at different temperatures. *Asian J. Chem.*, **22**: 1787-1798.
- Samanta A, Paul BK and Guchhait N (2011). Studies of bio-mimetic medium of ionic and non-ionic micelles by a simple charge transfer fluorescence probe N,N-dimethylaminonaphthyl-(acrylo)-nitrile spectrochimica acta Part A. *Molecular and Biomolecular Spectroscopy*, **78**: 1525.

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- Snehalatha T, Rajanna KC and Salprakash PK (1997). Methylene blue - ascorbic acid: An undergraduate experiment in kinetics. *J. Chem. Edu.*, **74**: 228.
- Uddin Fand Kazmi H (2003). A study of the shape of activated complex in reaction between potassium peroxodisulfate and potassium iodide. *Arab. J. Sc. Engg.*, **28**(2A): 136-144.
- Willson JG and Newall AB (1966). *General and Inorganic Chemistry*, Cambridge University Press, UK.