International Journal of ChemTech Research
CODEN (USA): IJCRGG ISSN: 0974-4290
Vol.7, No.01, pp 412-419, 2014-2015

Mechanistic studies on β-cyclodextrin catalyzed oxidation of glutamine

S. Shunmugakani[1] and D. Easwaramoorthy[2]*

[1]Department of Chemistry, Agni College of Technology, OMR,Thalambur, Chennai-603103, India.
[2]Department of Chemistry, B.S.Abdur Rahman University,Chennai-600 048, India.

Abstract: Kinetics and mechanistic study of β-cyclodextrin catalyzed oxidation of glutamine by peroxomonosulphate was investigated. It was found that the reaction was first order with respect to [glutamine] and [PMS]. Variation of the ionic strength and the solvent polarity had negligible effect on the rate of the reaction. Activation and thermodynamic parameters for the oxidation of glutamine were calculated. The interaction of β-cyclodextrin and glutamine was analyzed by UV- Vis spectrophotometer to determine the formation of inclusion constant and the constant obtained was 72.76 x10^3 M^{-1}.
Key words: Oxidation, glutamine, peroxomonosulphate (PMS), β-cyclodextrin.

1. Introduction

Cyclodextrins (CD) are cyclic oligomers of glucose that form guest- host complexes with many species including inorganic anions in solution [1]. Kinetics of cleavage of phenyl-phenyl acetates catalyzed by β cyclodextrin in basic aqueous sodium carbonate – bicarbonate buffer medium has been reported [2]. Kinetic study of the oxidation of amino acids and bovine serum albumin (BSA) by the free radicals in the absence or presence of α or β cyclodextrin was investigated [3]. The main interest in cyclodextrins lies in their ability to form inclusion complexes with several compounds [4–8]. EPR studies about interaction of CDs with flexible bi radicals were also reported [9-10]. The literature survey revealed that the kinetics and mechanistic study of effect of β cyclodextrin on the oxidation of glutamine by peroxomonosulphate was not reported in the literature and hence the title study was carried out and the results were presented here.

2. Experimental Methods

2.1 Materials and Reagents

PMS was obtained from Aldrich, USA, and the purity of the sample was found to be 98% when tested by iodometric estimation and hence used without further purification. PMS solution was freshly prepared every day, stored in a blackened vessel to prevent photodecomposition, and standardized iodometrically.

Glutamine was obtained from Merck, India, and used as received. β-cyclodextrin was purchased from SD-Fine chemicals, India. Acetic acid was distilled to remove impurities and used to make the buffer solution. Analar grade solvents such as acetonitrile and 2-methyl-2-propanol were distilled and used for the reactions.

2.2 Kinetic Measurements

The kinetics of oxidation of glutamine by PMS in the presence of β-cyclodextrin in acetic acid–sodium acetate buffered medium was studied under pseudo-first-order conditions, i.e., [glutamine] >> [PMS] .A known volume of PMS solution, thermostated at the desired temperature, was pipetted out into the reaction mixture and

simultaneously a timer was started. Consumption of PMS in this reaction mixture was monitored by iodometric method. The rate of the reaction followed first-order kinetics as shown in Fig. 1,

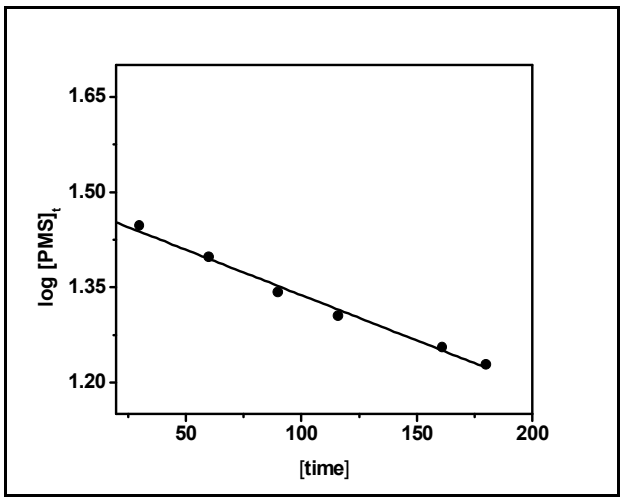


Fig.1 Plot of log $[PMS]_t$ vs. time at 308 K

Fig.1 Plot of log $[PMS]_t$ vs. time at 308 K
[Glutamine]= 0. 05 mol dm^{-3}; [sodium acetate] =0.085 mol $dm^{-3;}$ β Cyclodextrin = 0.3g
pH =4.0 ± 0.1; [PMS] =3.98×10^{-3} mol dm^3

The method of least squares was used to calculate the slope and intercept. The relative standard errors of the above-mentioned rate constant for a single run and the relative standard errors of the mean were about 4% and 2% respectively. The accuracy of the temperature was ± 0.1º C pH was measured using globe pH meter and the accuracy was ±0.1units

2.3 UV Spectral analysis

The reaction mixture was scanned in the ultraviolet and visible regions on Perkin Elmer LS 25 UV spectrophotometer absorption spectra were determined.

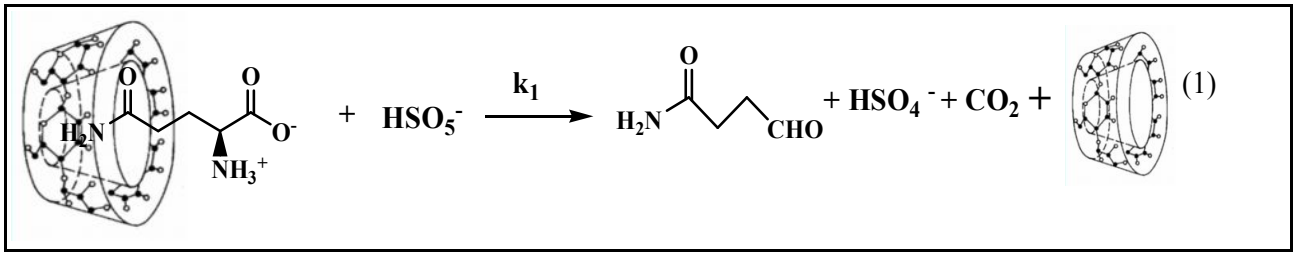
2.4 Cyclic voltammetric studies

The electrochemical studies were carried out using (CHI 760C – CH Instrument Inc., USA), three electrodes single compartment cell setup were employed for the electrochemical experiments. Here, glassy carbon, platinum wire, and Ag/AgCl electrode used as working electrode, counter electrode and reference electrode, respectively. All potentials were reported with respect to Ag/AgCl electrode.

3 Results and discussion

3.1 Stoichiometry

The stoichiometry of the reaction was determined for the reaction mixtures containing a large excess of [PMS] over [glutamine] .Then the reaction mixture was kept for 48 h and the unconsumed PMS was estimated iodometrically. Corrections for the self-decomposition of PMS was made from the value obtained from the control experiments. The observed stoichiometry of the reaction of β-cyclodextrin and glutamine: PMS was 1:1.



3.2 Product Analysis

The reaction mixture containing a large excess of PMS over glutamine in acetic acid- sodium acetate buffer was kept for 48 h in a blackened vessel at room temperature. Excess PMS present in the reaction mixture was destroyed by adding sodium bisulphite and then the mixture was extracted with dichloromethane. The organic layer was separated, dried and given for IR analysis.

3.3 Effect of [glutamine] on k_{obs}.

The values of k_{obs} were calculated for the reactions studied for different concentrations of [glutamine] by keeping all other parameters at constant values. Perusal of the kinetic results showed that the rate constant increased with increase in [glutamine] Table 1. Further, the plot of k_{obs} vs. [glutamine] was linear Fig. 2 This result indicated first order dependence of rate on glutamine. The positive intercept obtained in the above plots revealed that the reaction proceeded in two steps: one dependent on [glutamine] and the other independent of [glutamine]. The glutamine -independent step was due to the self-decomposition of PMS under the experimental conditions employed in this study.

Table 1: Effect of varying concentrations on the reaction rate at 308K

10^3 x [PMS] (mol dm^{-3})	10^2 x [Glutamine] (mol dm^{-3})	10^2 x [sodium acetate] (mol dm^{-3})	pH$\pm$ 0.1	β-Cyclodextrine (gram)	10^5 x k_{obs} (s^{-1})	Temperature (K)
1.93	5.00	8.50	4.0	0.3	13.80	308
3.86	5.00	8.50	4.0	0.3	6.90	308
5.79	5.00	8.50	4.0	0.3	5.30	308
7.72	5.00	8.50	4.0	0.3	4.90	308
3.86	0.025	8.50	4.0	0.3	0.23	308
3.86	0.0375	8.50	4.0	0.3	0.268	308
3.86	0.05	8.50	4.0	0.3	0.422	308
3.86	0.0625	8.50	4.0	0.3	0.498	308
3.86	5.00	2.13	4.0	0.3	3.84	308
3.86	5.00	4.25	4.0	0.3	3.84	308
3.86	5.00	6.38	4.0	0.3	3.61	308
3.86	5.00	10.63	4.0	0.3	3.21	308
3.86	5.00	8.50	3.6	0.3	3.45	308
3.86	5.00	8.50	4.0	0.3	6.9	308
3.86	5.00	8.50	4.4	0.3	0.537	308
3.86	5.00	8.50	4.8	0.3	14.58	308
3.86	5.00	8.50	4.0	0.1	0.422	308
3.86	5.00	8.50	4.0	0.2	0.46	308
3.86	5.00	8.50	4.0	0.3	0.498	308
3.86	5.00	8.50	4.0	0.5	0.537	308
3.86	5.00	8.50	4.0	0.7	0.580	308
3.86	5.00	8.50	4.0	1.0	0.69	308
3.86	5.00	8.50	4.0	1.5	0.729	308
3.86	5.00	8.50	4.0	0.3	0.422	303
3.86	5.00	8.50	4.0	0.3	0.69	308
3.86	5.00	8.50	4.0	0.3	0.7676	314
3.86	5.00	8.50	4.0	0.3	1.1515	318

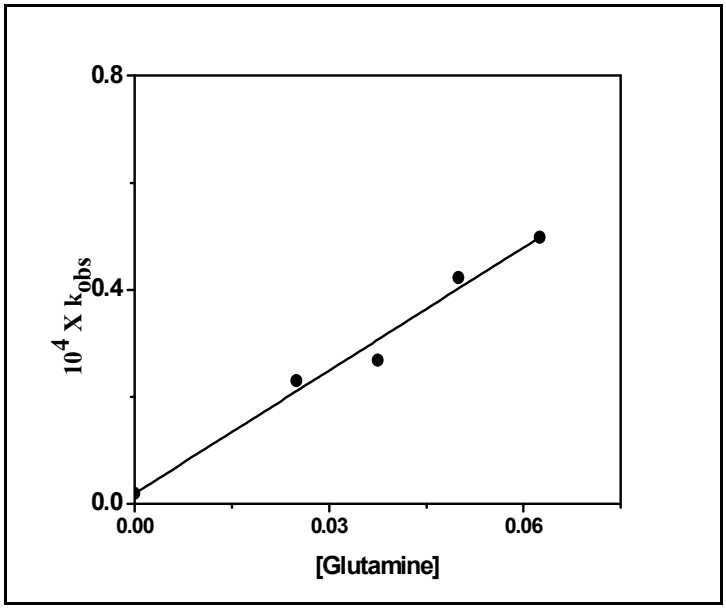


Fig.2 Plot of k_{obs} vs. [Glutamine] at 308K

Fig.2 Plot of k_{obs} vs. [Glutamine] 308 K
[sodium acetate] =0.085 mol dm^{-3}; pH =4.0 ± 0.1;[PMS] =3.98×10^{-3} mol dm-$^{3;}$
β Cyclodextrin=0.3g

3.4 Effect of pH on k_{obs}.

In order to ascertain the effect of pH on the rate, the reactions were carried out at different pH values. The rate constant (k_{obs}) values increased with an increase in pH values. The plot of k_{obs} vs. [$1/H^+$] gave a straight line.

3.5 Dependence of the rate on [β-CD]

The values of k_{obs} were calculated for the reactions conducted for different quantities of β-cyclodextrin by keeping the other parameters at constant values. The rate of a reaction increased with increase in [β-cyclodextrin] Table 1. The plot k_{obs} vs. [β-cyclodextrin] was linear with a positive intercept (Fig. 3).

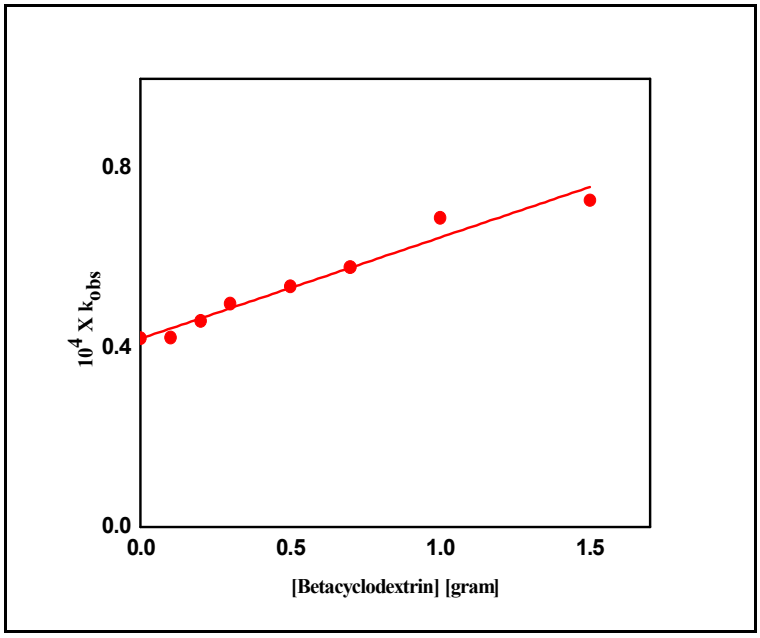


Fig.3 Plot of k_{obs} vs. [β-Cyclodextrin] at 308 K

Fig.3 Plot of k_{obs} vs. [β-Cyclodextrin] 308 K
[Glutamine]= 0. 5 mol dm^{-3}; [sodium acetate] =0.085 mol dm^{-3} pH =4.0 ± 0.1;
[PMS] =3.98×10^3 mol dm^{-3}

3.7 Effect of Temperature on k_{obs}

The reaction was studied at five different temperatures, viz., 303, 308, 313 and 318 K, by keeping all the other parameters constant. The k_{obs} increased with the increase in temperature, and the plot of log k_{obs} vs. 1/T was a straight line (Arrhenius plot). A plot of log k_{obs}/T vs. 1/T was also linear (Eyring's plot). From the slope and intercept of the straight line, the thermodynamic parameters were calculated (Table 2). The positive values of free energy of activation (ΔG) and enthalpy of activation (ΔH) obtained in this study indicated that transition state was highly solvated, while the negative values of entropy of activation (ΔS) suggested the formation of a rigid transition state than the reactants with reduction of degree of freedom of molecules.

Table 2: Kinetic and thermodynamic parameters for the oxidation of glutamine at 308 k

Glutamine	E_a kJ mol^{-1}	$\Delta H^{\#}$ kJ mol^{-1}	$\Delta S^{\#}$ J $K^{-1}mol^{-1}$	$\Delta G^{\#}$ kJ mol^{-1}
Glutamine	31.64	33.39	-149.14	47.91

3.8 Effect of Dielectric Constant on k_{obs}

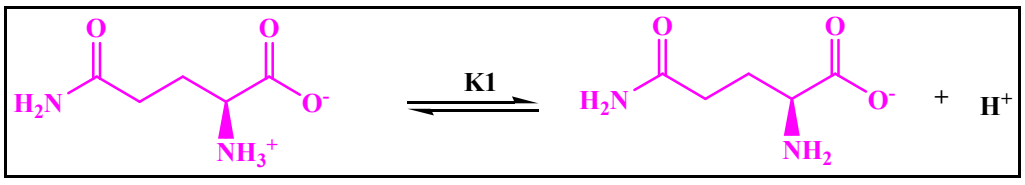
The effect of the dielectric constant (ε) of the reaction mixture on the reaction rate was studied by using two different solvents, such as 2-methylpropan-2-ol (tertiary butyl alcohol) and acetonitrile. The k_{obs} remained unaffected with the increase in composition of the solvents, ruling out the formation of a polar intermediate.

3.9 Effect of Ionic Strength on k_{obs}

The effect of ionic strength on the reaction rate was studied by varying the ionic strength of the medium from 0.05 to 0.2 mol dm^3 by maintaining the other parameters at constant values. The increase in the ionic strength had negligible effect on the k_{obs} value. The absence of any effect of ionic strength on k_{obs} revealed that HSO_5^- preferably attacked the amino group rather than the carboxylic acid group of glutamine.

Discussion

Glutamine exists as a dipolar ion in aqueous solutions. The dissociation of glutamine depends on the pH of the medium. The pKa value suggested that in acidic medium, glutamine exist both in the protonated form and as zwitterions as shown below



The formation of inclusion complex was confirmed by UV-Visible absorption studies. This can be done by varying the concentration of guest (glutamine) 50 mg and fixed concentration of host (β-cyclodextrin) 500 mg. It was observed that the intensity decreased due to the formation of inclusion complex. The stability constant was calculated by plotting 1/A vs. [1/ β-CD] (Benesi-Hildebrand equation). The plots of 1/A vs. [1/ β-CD] was linear as shown in (Fig.5) with very good linear relationship with R^2=0.993 and the stability constant value was 72.76 x10^3 L/ mol. . The stoichiometry ratio for the inclusion complex formation between glutamine and β-CD was 1:1

$$\frac{1}{A} = \frac{1}{\varepsilon [G]_0 K[CD]} + \frac{1}{\varepsilon [G]_0} \qquad (3)$$

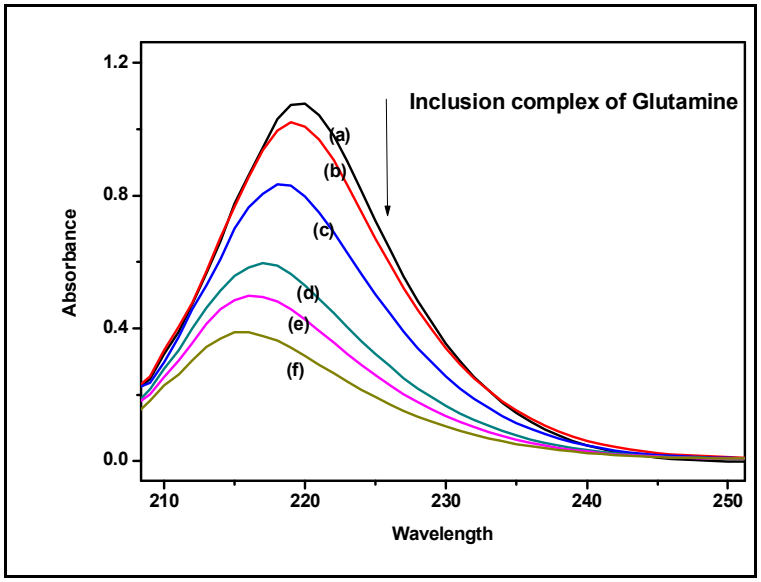


Fig.4 Absorption spectra of glutamine with various concentration of β-Cyclodextrin

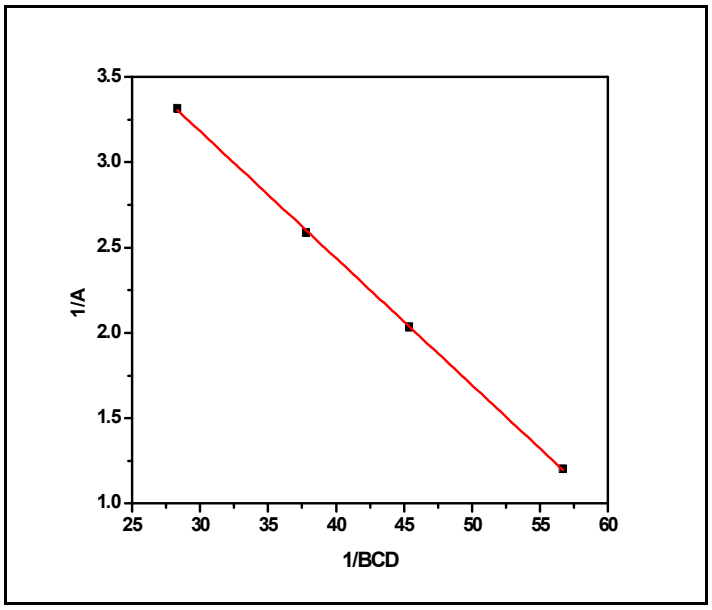


Fig.5 Plot for 1/A against 1/[β–Cyclodextrin] of glutamine inclusion complex.

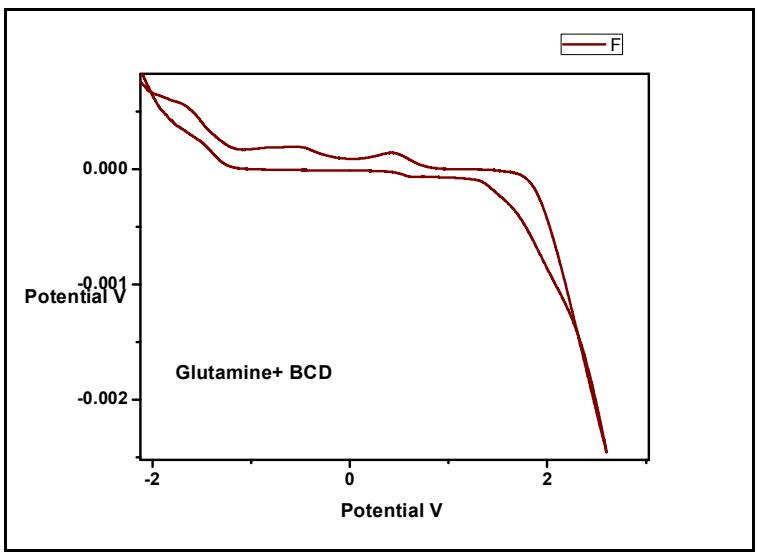
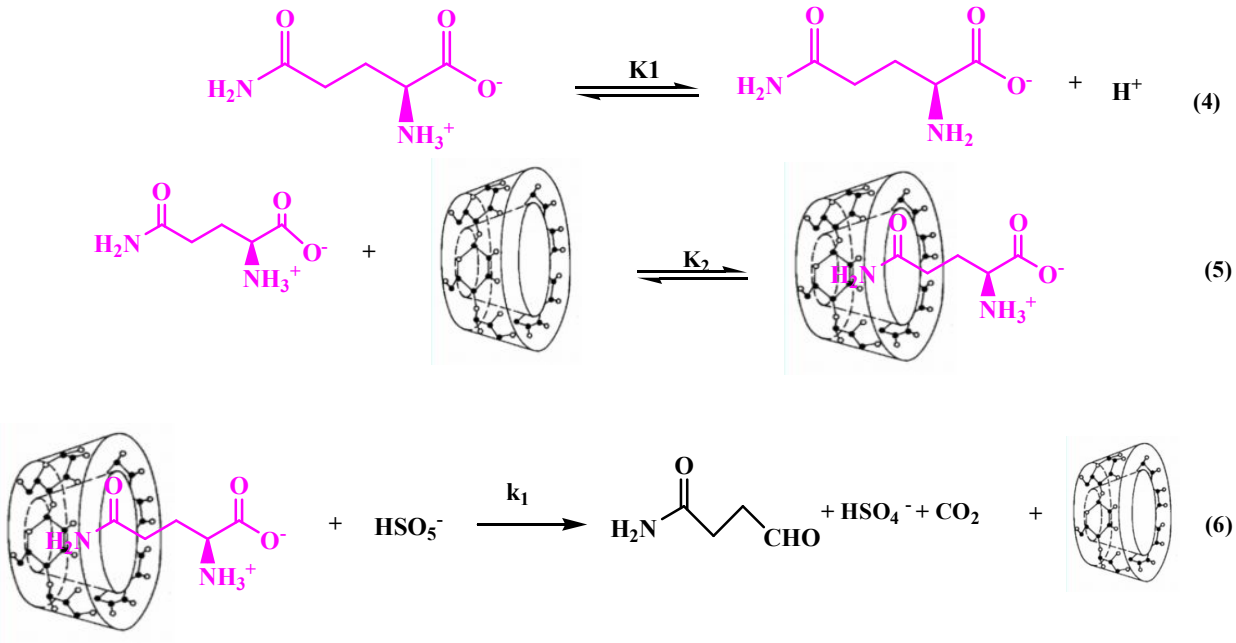


Fig. 6 Cyclic voltammogram of the reaction mixtures of Buffer +Glutamine + β Cyclodextrin

Scheme1



$$\frac{-d\,[HSO_5^{--}]}{dt} = k_1\,[\text{complex}]\,[HSO_5^-] \qquad (7)$$

$$= \frac{k_1 K_1 K_2\,[\text{Glutamine}]\,[BCD]}{K_1 + [H^+]} \qquad (8)$$

$$k_{obs} = \frac{k_1 K_1 K_2\,[\text{Glutamine}]\,[BCD][HSO_5^-]}{K_1 + [H^+]} \qquad (9)$$

$$= \frac{k_1 K_1 k_2\,[\text{Glutamine}][BCD]}{K_1 + [H^+]} \qquad (10)$$

5. Conclusion

The Kinetics of the oxidation of glutamine by PMS in acetic acid–sodium acetate buffered medium (pH 3.6–5.2) using β-CD as catalyst was studied at 308 K. The reaction was carried out at four different temperatures, and the activation and thermodynamic parameters were calculated. A suitable reaction mechanism was proposed to explain the experimental observation. Cyclic voltammetric studies and absorption studies confirmed the formation of inclusion complex. Based on the above discussion, the detailed mechanism was proposed.

Acknowledgement:

The authors acknowledge the authorities of B.S. Abdur Rahman University, Chennai-600 048, India. The author, S. Shanmugakani acknowledges the principal and the management of Agni College of technology, OMR, Thalambur.Chennai-600 130, India, for their constant encouragement and support.

References

1. Szejtli J. Chem. Rev.1998, 98 5, 1743.
2. Raj V, Chandrakala T, Rajasekaran K , J.Chem. Sci, 119, 2008, 327-328
3. Ionita *G,* Ionita P*,* Sahini V *,* Luca C*,* J. Incl. Phen. Macrocyclic Chem *, 39 ,2001, 269-271*
4. Hedges RA. Industrial applications of cyclodextrins. Chem, Rev 1998, 98, 2035–44.
5. Lu X, Chen Y. Chiral separation of amino acids derivatized with fluoresceine-5-isothiocyanate by capillary electrophoresis and laserinduced fluorescence detection using mixed selectors of beta-cyclodextrin and sodium taurocholate. J Chromatogr A 2002,955,133–40.
6. Baudin C, Pean C, Perly B, Goselin P. Inclusion of organic pollutants in cyclodextrin and derivatives. Int Environ Anal Chem, 2000, 77,233–42.
7. Kumar R, Dahiya J S, Singh D, Nigam P. Biotransformation of cholesterol using Lactobacillus bulgaricus in glucose controlledbioreactor.Bioresour Technol ,2001,28, 209–11.
8. Koukiekolo R, Desseaux V, Moreau Y, Marchis MG, Santimone M. Mechanism of porcine pancreatic alpha-amylase inhibition of amylose and maltopentaose hydrolysis by alpha-, beta- and gammacyclo dextrins. Eur J Biochem ,2001, 268, 841–8.
9. Gagnaire G, Michon J, Pierre J. L, New J. Chem., 1992, 16, 915.
10. Ionita G ,and Chechik V , Org. Biomol. Chem, 2007, 5, 1910.

*****