PRECONCENTRATION OF SUDAN I DYE USING B-CYCLODEXTRIN EPICHLOROHYDRIN POLYMER AS THE SOLID PHASE EXTRACTANT

Dr. Rupinder Kaur* and Dr. Ashok Kumar Malik

Department of Chemistry, Punjabi University, Patiala, Punjab, India.

*Corresponding Author
Dr. Rupinder Kaur
Department of Chemistry,
Punjabi University, Patiala,
Punjab, India.

ABSTRACT

In this study, a solid phase extraction method has been developed for the preconcentration of Sudan I dye at the trace level using β-Cyclodextrin polymer. After preconcentration the dye solute was determined UV-VIS spectrophotometry. Various parameters, such as effect of pH, sample volume, shaking time, amount of adsorbent, agitation speed for the % uptake of Sudan I has been optimized. This method has been applied for the determination of Sudan I in different food samples.

KEYWORDS: β-Cyclodextrin epichlorohydrin polymer, Sudan I dye, preconcentration, Spectrophotometry.

INTRODUCTION

Dyes play a significant role in food industry as food quality and flavor are closely associated with color of food. Dyes have been the subject of much interest in recent years because of increasingly strict restrictions on the organic content of industrial effluents. Food colorant or food dyes are both used in commercial food production and in domestic cooking. Sudan I is a synthetic azo dye, and is used in many fields such as paint or textile colorants, and floor or shoe polishes. Sudan I can cause tumours in the liver and bladder of mice, and is a possible human carcinogen and mutagen[1], therefore it has been classified as the third category carcinogen by the International Agency for Research on Cancer (IARC). Sudan I is forbidden as an additive of foodstuffs in any national and international food regulation act. In some places this harmful dye was used as an additive of foodstuffs to boost up the typical reddish color of chilli powder for commercial benefits. Therefore it is required to develop accurate

and fast methods to identify Sudan I dye in foodstuffs. Several methods has been proposed to detect dyes in food stuffs, such as liquid chromatography-photodiode array detection[2-3], molecularly imprinted solid phase extraction[4], and plasmon resonance light scattering.[5] Common methodologies include HPLC with ultra violet detection[6], enzyme-linked immunosorbent assay[7], atmospheric pressure chemical ionization-tendem mass spectrometry.[8] Some methods are very costly and are required the expert hands. So, spectrophotometry is widely used for the determination of dyes due to its higher sensitivity, low cost, low interference level and its excellent detection limits. So, brilliant green dye has been determined by spectrophotometric methods after preconcentration using β-CDP in food samples.

Supramoleculer complexes with β- cyclodextrin has been a very active research field in the past few years.[9-10] β- cyclodextrin (β-CD) is a very stable oligosaccharide that is composed of seven glucose units linked with each other by α-(1,4)-glycosidic linkage. It can form supramoleculer complexes with several organic compounds by incorporating them into their hydrophobic cavities. Two or more β-cyclodextrin covalently linked with each other are known as polymers. These β- cyclodextrin polymer have been used for the preconcentration of various analytes.[11-14] In the present work, β-cyclodextrin epicholorohydrin polymer (β-CDP) has been used as a solid support for the preconcentration of Sudan I dye.
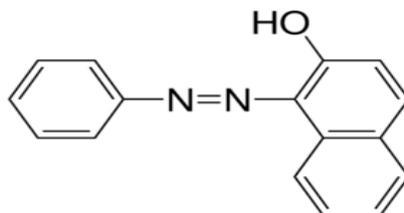
Fig. 1: Structure of 1- (phenyldiazenyl) napthalen-2-ol (Sudan I Dye).

Experimental

A Shimadzu UV-1800 spectrophotometer (Shimadzu Ltd., Japan) equipped with the matched 10 mm quartz cells was used to measure absorbance. Digital century pH-meter Cp - 901 with a combined glass electrode was used to carry out pH measurements. A thermostatic shaking water bath (Perfit India Ltd.) was used to carry out all the inclusive procedures.

Reagents

All chemicals used were of AnalR grade unless otherwise stated. Double distilled water was

used throughout the experiment. Brilliant green dye solution was prepared by dissolving 0.248 g in 100 mL of double distilled water to give 0.01 M standard stock solution and further diluted as and when required. β-CDP was synthesized by method given in Literature (1[a]). A brief procedure is given here : 40g of β-CD, 10 g of soluble starch and 100 mL of 20% sodium hydroxide were added into a beaker. The mixture was vigorously stirred at 50-60ºC until the reactants dissolved. Total 60 mL of epichlorohydrin was added drop wise into the solution, and β-CDP was formed in 30 min. Filtered with pressure through Buchner funnel and then washed with distilled water 5-6 times, the polymer was dried at 100ºC and then stored at room temperature in the desiccators for further use.
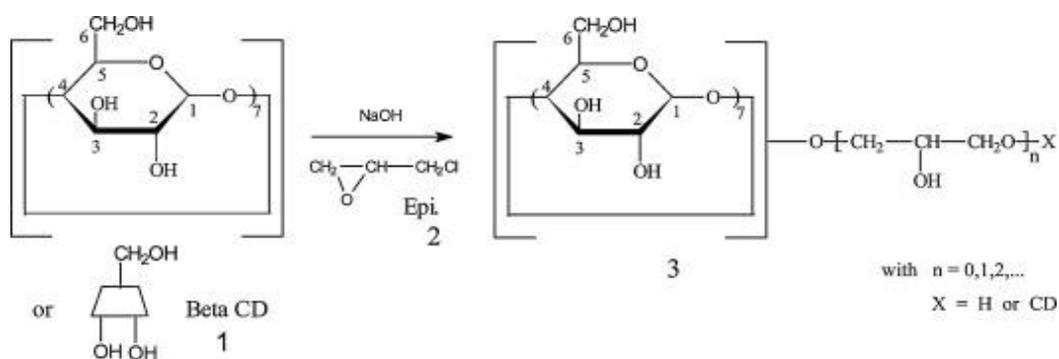
Fig.2: Schematic representation of the synthesis of β-cyclodextrin polymer.

The synthesized polymer was subjected to SEM analysis to study their morphological aspects. SEM Scans of the polymers were carried out at Sophisticated Instrumentation centre (SIC), Punjabi University, Patiala on JEOL (Tokyo, JAPAN) JSM-6510/LV-SEM Instrument. For the above experiment 1mg. of the sample was mounted on a glass slide and subjected to scan at an acceleration voltage of 10Kv. The SEM images were taken at three different magnifications from X200, X1000 and X5000. From the SEM micrographs it is obvious that the structure of β-CD is rock like and β-CDP is relatively fibrous which shows that cross-linking has taken place.
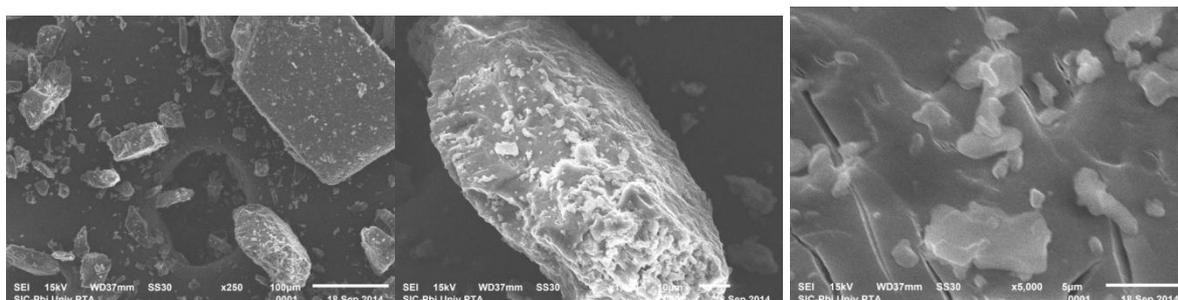
Fig. 3: SEM scan of β-CD at (a) X200, (b) X1000, (c) X5000.
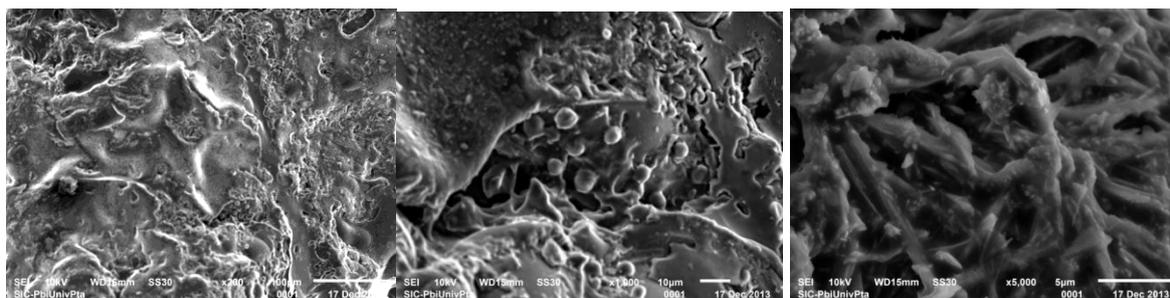
Fig. 4: SEM scan of β-CDP at (a) X200, (b) X5000, (c) X5000.

Buffer solution in the pH range of 2.0-3.5 were made by mixing equimolar solutions of hydrochloric acid/Sodium Acetate and buffer solutions in the pH range of 4.0-6.5 were made by mixing equimolar solutions of sodium acetate and acetic acid solutions in the different proportions While those in the pH range of 7.0-11.0 were made by mixing equimolar solutions of ammonia and ammonium chloride. The glass wares were washed with chromic acid and soaked in 5% nitric acid and then cleaned with double distilled water before use and dried in an electric oven.

Procedure

200 mg of β-CDP and 2.5 mL of buffer solution (pH 3.0) were added to a 100 ml stoppered conical flask at room temperature. The mixture was allowed to stand for 15 min. so that β-CDP should swell sufficiently and appropriate amount of dye was added and made up to 75 ml with double distilled water. The mixture was shaken in the thermostatic shaking water bath for 90 min. at the rate of 100 r.p.m., 5.0 ml of supernatant solution was transferred into a test tube and the absorbance was measured spectrophotometrically.

Optimization of various parameters

Effect of pH

The formation of inclusion complex of the dye in the polymer depends on the pH of the sample solution which was studied in the range of (1.0-7.0) using different buffer solutions, % uptake ($\geq$ 95) was obtained at pH 3.0. (Fig.5). Therefore, the working pH was chosen as 3.0 for the subsequent studies.
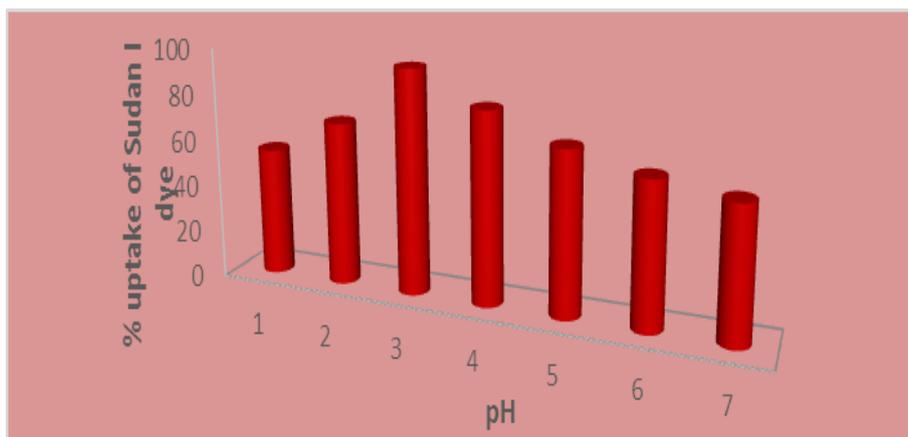
Fig.5: Effect of pH on the % uptake of the Sudan I dye by the β-CD polymer.

Effect of shaking time

Shaking time is an important factor in determining the possibility of application of the β-CD polymer for the selective uptake of Sudan I dye. Different shaking time (ranging from 15 to 120 min) were studied for the % uptake of Sudan I dye by β-CD polymer. The results of % uptake of Sudan I dye vs. the shaking time show that the % uptake of (≥95%) was attained in 90 min.(Fig.6). Therefore, the shaking time of 90 min. was selected for further studies.
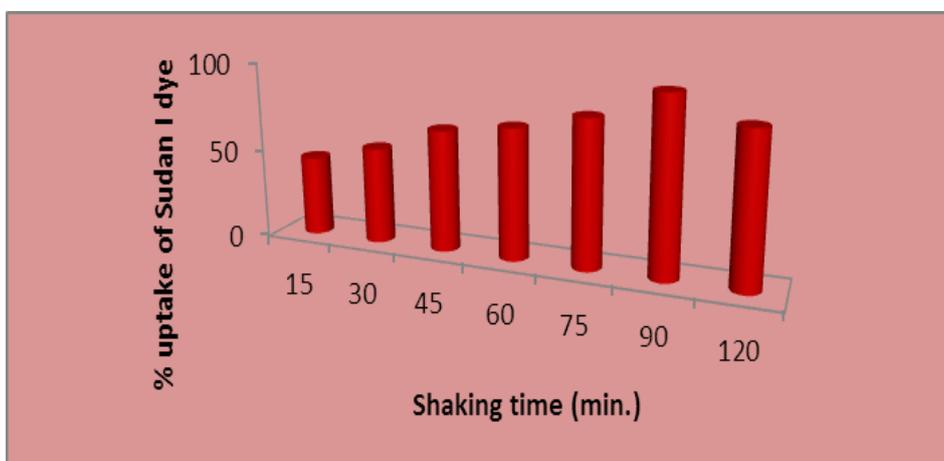
Fig.6: Effect of shaking time on the % uptake of the Sudan I dye by the β-CD polymer.

Effect of sample volume

Enriching low concentration of dye from large volume of sample the effect of sample volume is an important factor in determining the possibility of application of polymer for the % of uptake of Sudan I dye.For this purpose 15, 30, 45, 60, 75 and 90 mL of sample volumes containing a fixed amount of dye were taken and uptake of Sudan I dye was studied (Fig.7). The maximum % uptake (≥95%) of Sudan I dye was obtained at sample volume of 75 mL. Therefore, 75 mL of sample volume was used for further studies.
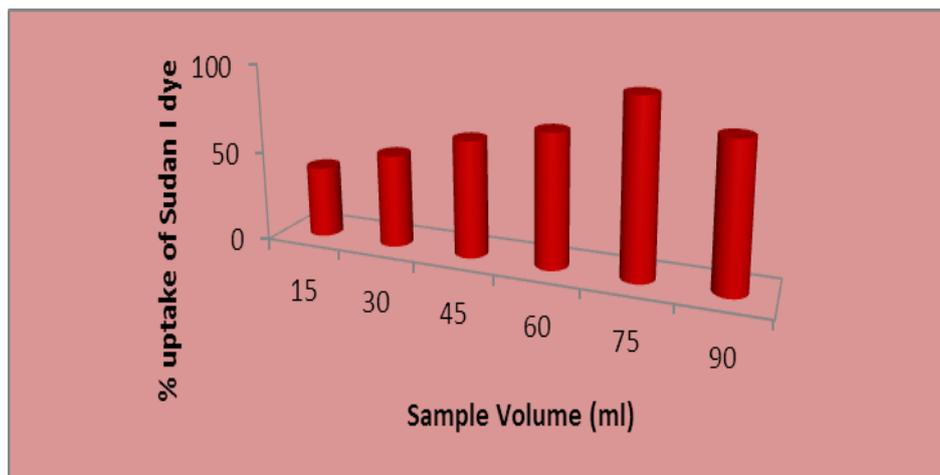
Fig.7:Effect of sample volume on the % uptake of the Sudan I dye by the β-CD polymer.

Effect of agitation speed

Speed of shaking is an important factor in determining the possibility of application of polymer for the quantitative % uptake of Sudan I dye. The driving force i.e Shaking speed could help in mass transfer and facilitate the concentration gradient between the sample solution and the polymer. Different speeds (ranging from 40 to 140 r.p.m) were studied for the % uptake of Sudan I dye by the polymer. The results of % uptake of Sudan I vs. agitation speed (Fig. 8) show that the % uptake reach maximum (≥95%) at 100 r.p.m. Therefore, the shaking speed of 100 r.p.m. was selected for further studies.
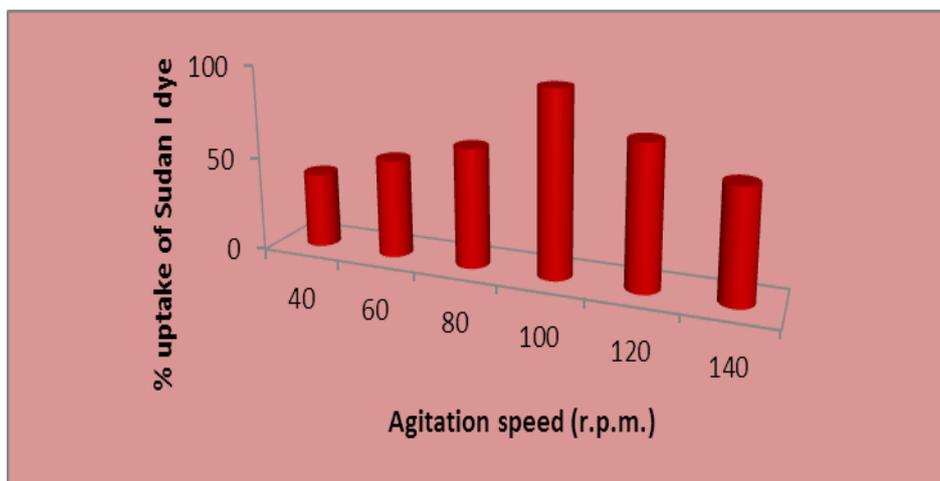
Fig.8: Effect of agitation speed on the % uptake of the Sudan I dye by the β-CD polymer.

Effect of amount of polymer

The amount of the β-CD polymer is another important parameter that affects % uptake of dye. A quantitative removal (≥ 95%) cannot be achieved when the β-CD polymer is less than

the optimum amount. In order to optimize the smallest amount of polymer, 100 mg, 200 mg, 300 mg, 400 mg and 500 mg of the polymer were added to the solution containing known amount of dye. The quantitative recoveries were obtained at 200 mg of β-CDP (Fig.9). Therefore, 200 mg of the β-CDP has been used for further studies.
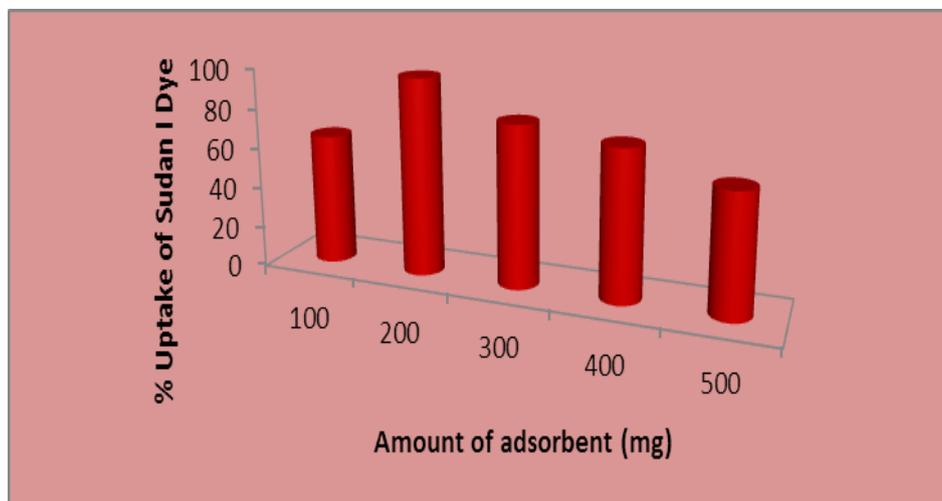
Fig.9: Effect of amount of adsorbent on the % uptake of the Sudan I dye by the β-CD polymer.

Applications

Determination of samples

The proposed method has been applied for the determination of Sudan I dye in Chilli Sauce and Tandoori masala mix. The results are given in table.

Table 1. Result of determination of Sudan I in food samples

Food Samples	Added, µg/ml	Found, µg/ml	Recovery, %
[a]Tandoori masala mix	0	0.006	-
	0.350	0.348	97.75
	0.700	0.698	98.86
	0.932	0.930	99.14
	0	0.015	-
[b]Chilli Sauce	0.331	0.332	95.95
	0.662	0.650	96.01
	0.892	0.895	98.45

[a]Tandoori masala mix, [b]Chilli Sauce – locally avaiable in market.

CONCLUSION

The proposed preconcentration method consist of a simple and low procedure which permits the quantitative recovery of Sudan I dye from food samples. The synthesis of the polymer is

easy and the method has a good accuracy, sensitivity and repeatability. The polymer has been used in all the experiments performed for the study. It has a unique stability and reusability. This method is convenient for the determination of Sudan I dye.

REFERENCES

1. Stiborova, M.; Martinek, V.; Rydlova, H.; Hodek, P.; Feri, E.; *Cancer Res*, 2002; 62: 5678.

2. Zhang, Y.; Wu, H.L.; Xia, A.L; Han, Q.J.; Cui, H.; Yu, R.Q.; *Talanta*, 2007; 72: 926.

3. Mazzetic, M.; Fascioli, R.; Mazzonci, I.; Spinelli, G.; Morelli, I.; Bertoli, A.; *Food Addit. Contam*, 2004; 21: 935.

4. Puoci, F.; Garreffia, C.; Lemma, F.; Muzzalupo, R.; Spizzirri, U. G.; Picci, N.; *Food Chem*, 2005; 93: 349.

5. Wu, L.; Li, Y.; Huang, C.; Zhang, Q.; *Anal. Chem,* 2006; 78: 5570.

6. Cornet, V.; Govaert, Y.; Moens, G.; Loco, Van. J.; Degroodt, J.M.; *J. Agri. Food. Chem*, 2006; 54: 639.

7. Wang, Y.; Wei, D.; Yang, H.; Yang, Y.; Xing, W.; Li, Y.; Den, A.; *Talanta*, 2009; 77: 1783.

8. Donna, L. Di.; Maluolo, L.; Mazzatti, F.; De, Luca. D.; Sindona, G.; *Anal Chem*, 2004; 76: 5104.

9. Li, R.; Jiang, Z. T. and Liu, Y. H.; *J Food Drug Anal,* 2008; 16(5): 91-96.

10. Velic, D.; Knapp, M. and Kohler, G.; *J Mole Stucture*, 2001; 598(1): 49-56.

11. Abay, I.; Denizli, A.; Biskin, E. and Salih, B.; *Chemosphere,* 2005; 61(9): 1263-1272.

12. Shao, D.; Sheng, G.; Chen, C.; Wang, X. and Nagastu, M.; *Chemosphere,* 2010; 79(7): 679-685.

13. Wu, M. and Zhu, X.; *Spectrochim Acta A Mol Biomol Spectrosc.*, 2010; 77(5): 1021-1024.

14. Bhaskar, M.; Aruna, P.; Rama, Jeevan.; Ganesh, Jeevan.; and Radhakrishnan, G.; *Anal Chim Acta,* 2004; 509(1): 39-45.