

Photolytic oxidation of Coomassie Brilliant Blue with H₂O₂

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Received 12 May 2004; received in revised form 21 June 2004; accepted 10 September 2004

Available online 8 December 2004

Abstract

Coomassie Brilliant Blue, a biochemically important dye, was subjected to UV radiation in the presence of hydrogen peroxide. The photo-oxidation of the dye was monitored spectrophotometrically. The rate of decolorisation and degradation were calculated from the observed data and were found to be first order. A plausible explanation involving the probable radical initiated mechanism was given to explain this behaviour.

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Keywords: Coomassie Brilliant Blue; Photolysis

1. Introduction

Most of the dyes such as azo dyes are used in industrial processes, like in the leather, textile and printing industries. Some of these dyes are toxic in nature and their removal from the industrial effluents is a major environmental problem [1–4]. Untreated effluents are highly coloured and thus particularly objectionable if discharged to open waters. The dye concentration of the effluent may be well below the desired limits, but the dye will impart colour even at very low concentrations. Incomplete discoloration of the effluent prior to discharge makes the water treatment process difficult and these dyes often end up as sludge, which are then removed and deposited in landfills. Quite apart from the aesthetic desirability of coloured streams resulting from dye waste, azo dyes in particular can undergo anaerobic degradation to potentially carcinogenic amines [5]. Literature review on this subject matter has revealed the importance of photo catalytic and photolytic degradation of various dye solutions [6–12].

Coomassie Brilliant Blue, a non-azo dye, which was originally designed to be used as an acid wool dye, is an extremely popular reagent for protein chemists. This dye is used extensively for staining proteins in electrophoresis techniques, as well as for measuring protein concentrations (e.g. Bradford protein assay). The purpose of this study is to focus attention on the photolytic degradation of Coomassie Brilliant Blue, a non-azo dye, in the presence of H₂O₂ and UV light and investigate the kinetic parameters.

2. Experimental

Coomassie Brilliant Blue (F.W.=854) with a labeled purity of more than 90% was obtained from Biorad (USA) and used as such. Deionised water was used to make the dye solutions of desired concentration. Hydrogen peroxide (35% w/w) was obtained from Merck and used in this work. Its concentration in all the dye solutions for this work was kept constant at 10 mM. UV/VIS studies were done on a CARY 50 UV/VIS spectrophotometer, using a 1 cm quartz cell. For photolytic experiments, the samples were irradiated in a UV box procured from Kodak. The instrument has

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a series of four UV lamps set in parallel which operate at 25 W with a UV output at 302 nm in both the high and low settings. In the present work, the instrument was used in the low output mode unless otherwise mentioned. The pH of the solution was measured by using a digital pH meter.

2.1. Preparation of samples and decoloration/ degradation studies

Coomassie Brilliant Blue stock solution of 1.2×10^{-3} M was prepared in 100 mL of deionised water in a 250 mL flask by first dissolving the dye in methanol and then adding glacial acetic acid to a final concentration of 0.5% and methanol at 20%. Necessary dilutions of this stock were done with deionised water. Unless otherwise indicated, the working concentration of the Coomassie Brilliant Blue dye was 24 μ M. A 50 mL aliquot of this diluted solution (24 μ M) was taken and 48.5 μ L of H_2O_2 was added to this. The concentration of H_2O_2 in this solution is estimated to be 10 mM. The pH of the solution was found to be 3.14. No attempt was made at this stage to study the effect of pH on the discoloration of the dye. This effect along with the effect of other additives will be reported in another study. The contents were irradiated with a UV light of 302 nm for a given period of time. A series of such solutions were taken in different tubes and subjected simultaneously to UV radiation. After a certain time interval, one tube was drawn out from the UV box and the absorbance of the solution was monitored instantaneously on a spectrometer. The absorbance value obtained in each case was plotted against time to obtain the order of discoloration rate. Photolytic oxidation studies were carried out at 25 ± 2 °C.

3. Results and discussion

In the present work, the kinetics of H_2O_2 assisted photochemical oxidation of Coomassie Brilliant Blue was investigated. The structure of the dye is given in Fig. 1. The initial investigation of the dye revealed two

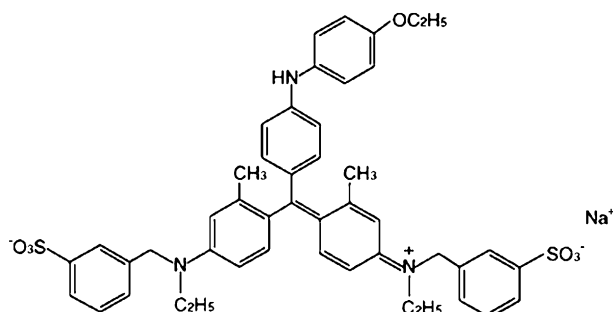


Fig. 1. Structure of Coomassie Brilliant Blue G-250.

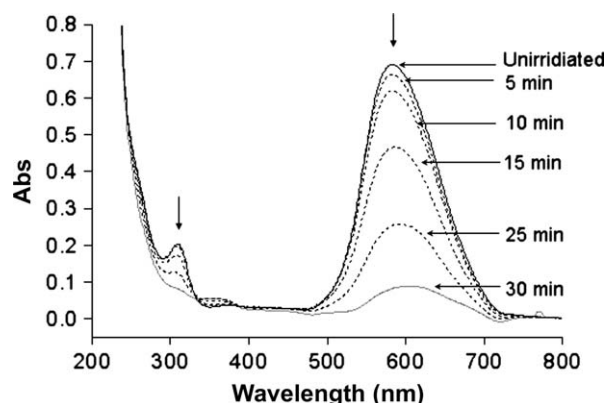


Fig. 2. Time-dependent photolytic oxidation of Coomassie Blue dye G-250 with different times of irradiation. [Dye]=24 μ M, [H_2O_2]=10 mM.

main peaks in its absorption spectra in aqueous solution, one at 585 nm and the other at 312 nm as shown in Fig. 2. These can be assigned to $n \rightarrow \pi^*$ transition (attributed to the colour of the dye) and the $\pi \rightarrow \pi^*$ transition (attributed to the delocalised aromatic structure of the dye), respectively. For decoloration studies, 585 nm wavelength was chosen for further investigations, whereas for degradation studies, 312 nm wavelength was targeted.

The degradation of this dye in the presence of H_2O_2 with UV light was carried out. Deionised water was used as a reaction media. Initially experiments were carried out in the absence and presence of either UV light or H_2O_2 alone. The results showed that mere UV light or H_2O_2 alone was not sufficient for degradation of this dye.

A solution of 24 μ M of the dye in water was prepared and subjected to UV light in the presence of H_2O_2 . The dye started decolorising immediately in the presence of H_2O_2 and the UV radiation. The rate of decolorising was monitored with respect to the decrease in absorption value of the dye solution, which has a distinct peak at 585 nm in the visible region. The decrease in the absorption spectra of the dye solution was monitored at regular intervals of time. Percentage decrease in absorption was calculated as follows:

$$\% \text{ decrease in absorption} = \frac{[A(\text{initial}) - A(\text{final})]}{A(\text{initial})} \times 100$$

The evolution of absorption for the decoloration and degradation of the dye solution as a function of irradiation time is given in Fig. 3. The effect of both types of UV intensities on dye discoloration and degradation was also investigated. It was shown that the % decolorisation was more with high intensity as compared to low intensity UV. For comparison purposes, the decolorisation at low intensity was found to be 10%, whereas at high intensity this value was 22%

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