APPLICATION OF EXPERIMENTAL DESIGN FOR PHOTODEGRADATION OF ROSE BENGAL (ACID RED 94)

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BORANG PENGESAHAN STATUS TESIS

JUDUL : **APPLICATION OF EXPERIMENTAL DESIGN FOR PHOTODEGRADATION OF ROSE BENGAL (ACID RED94)**

SESI PENGAJIAN: **2008/2009**

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APPLICATION OF EXPERIMENTAL DESIGN FOR PHOTODEGRADATION OF ROSE BENGAL (ACID RED 94)

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A thesis submitted in fulfillment
of the requirements for the award of the degree of
Bachelor of Chemical Engineering (Biotechnology)

Faculty of Chemical & Natural Resources Engineering
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April 2009

I declare that this thesis entitled "Application of Experimental Design for Photodegradation of Rose Bengal (Acid Red 94)" is the result of my own research except as cited in references. The thesis has not been accepted for any degree and is not concurrently submitted in candidature of any other degree.

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Name : Muhamad Zulhelmi Bin Mohamad Fithol

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Special Dedication to my beloved mother; Aminah Binti Din
and my hardworking father; Mohamad Fithol Bin Abdullah,

For all your endless care, support and trust in me.

ACKNOWLEDGEMENT

First and foremost, I would like to thank God for bestowing His grace and bringing wisdom on me until I finish my reseach. Next, I wish to express my biggest gratitude towards my thesis supervisor, Mr. Rozaimi Bin Abu Samah for his valuable guidance and support throughout my research. My appreciation also goes to my fellow friends for your cooperation and encouragement.

I am very thankful to Universiti Malaysia Pahang (UMP) for providing good facilities regarding my research in the campus. To all the technical unit staff in Faculty of Chemical & Natural Resources Engineering, thank you for your instructions and guidance.

My sincere appreciation also extends to all my fellow colleagues and others who have provided assistance at various occasions. Thank you for the time sacrificed to accompany me. And last but not least, I am grateful to all my family members. Without their constant encouragement and support, I would not have the concentration on doing a good job.

I hope this research will give the readers some insight as to the application of experimental design as well as dyes decolorization studies.

ABSTRACT

Advance Oxidation Process (AOP) of Acid Red 94 (AR 94) by UV and H_2O_2 system were carried out in this study. AR 94 was irradiated with UV light in the presence of H_2O_2. The photodegradation process of the dye was monitored spectrophotometrically. Effects of AR 94 and H_2O_2 concentrations, pH, and irradiation time for photodegradation of AR 94 were investigated throughout this research while other experimental conditions were fixed at specific values. Statistical approach was employed to study the effect of selected parameters with the aid of Design-Expert® 7.1.6 software. Two level factorial design was employed for the experimental design. From the result, it is shown that the highest percent of AR 94 degradation can be achieved was 92.31%. From the analysis of variance, it is found that the AR 94 concentration, pH, and time were significant factors along with the interaction factors of AR 94 concentration, H_2O_2 concentration, and pH which gave significant effect for AR 94 degradation as well. Then, the optimization process was done using response surface methodology (RSM). Based on ANOVA result, the proposed model can be used to navigate the design space. It was found that the response of AR 94 decolorization is very sensitive to the independent factor of pH. The proposed model for central composite design fitted very well with the experimental data with R^2 and R^2_{adj} correlation coefficients of 0.976 and 0.943, respectively. Analysis of results data shown that the optimum conditions suggested by the design of experiment were; 20μM AR 94, 0.05 M H_2O_2, 3.75 pH value and irradiation time 30 minutes.

ABSTRAK

Proses pengoksidaan *Acid Red 94* (AR 94) oleh sistem UV dan H_2O_2 telah dijalankan di dalam kajian ini. Radiasi cahaya UV dengan kehadiran H_2O_2 telah dikenakan ke atas AR 94. Proses fotodegradasi bagi AR 94 dipantau secara spektrofotometrik. Kesan-kesan kepekatan AR 94 dan H_2O_2, pH, dan masa radiasi untuk fotodegradasi AR 94 telah diselidik sepanjang penyelidikan ini manakala faktor-faktor eksperimen lain telah ditetapkan pada nilai-nilai yang khusus. Pendekatan statistik telah digunakan untuk mengkaji kesan terhadap kesan-kesan yang ingin dikaji dengan bantuan perisian Design-Expert®7.1.6. Rekabentuk faktoran dua-faktor telah dipilih dalam merekabentuk eksperimen. Hasil eksperimen menunjukkan, peratus degradasi AR 94 tertinggi yang boleh dicapai ialah 92.31%. Daripada analisa varians, didapati kepekatan AR 94, pH, dan masa radiasi adalah faktor yang penting di samping interaksi faktor antara kepekatan AR 94, kepekatan H_2O_2, dan nilai pH juga memberi kesan terhadap degradasi AR 94. Kemudian, proses pengoptimuman dibuat menggunakan Metodologi Permukaan Sambutan. Berdasarkan hasil dari ANOVA, model yang dicadangkan boleh digunakan untuk memanipulasi rekabentuk ruang. Hasil eksperimen mendapati bahawa respon fotodegradasi AR 94 sangat sensitif terhadap faktor tak bersandar pH. Model yang dicadangkan dengan Rekabentuk Gubahan Memusat juga sesuai dan selari dengan data eksperimen dengan nilai korelasi koeffisien R^2 and R^2_{tlrs} masing-masing 0.976 dan 0.943. Analisis bagi data hasil eksperimen menunjukkan keadaan optimum yang dicadangkan oleh aplikasi rekabentuk eksperimen adalah; 20µM AR 94, 0.05 M H_2O_2, nilai pH 3.75, dan 30 minit masa radiasi.

TABLE OF CONTENT

LIST OF TABLES

LIST OF FIGURES

LIST OF SYMBOLS/ABBREVIATIONS

ANOVA	-	Analysis of variance
AR 94	-	Acid Red 94
H_2O_2	-	Hydrogen Peroxide
min	-	minutes
UV	-	Ultraviolet
R^2	-	Regression correlation
μM	-	micromolar
M	-	molar
AOP	-	Advanced Oxidation Process
%	-	percentage
°C	-	degree Celsius
RSM	-	response surface methodology
C. I number	-	colour index number
HCl	-	hydrochloric acid
NaOH	-	sodium hydroxide
Sqrt	-	square root
nm	-	nanometer
R^2	-	regression correlation/korelasi koeffisien
R^2_{adj}.	-	adjusted regression correlation
R^2_{tlrs}	-	korelasi koeffisien terlaras

CHAPTER 1

INTRODUCTION

1.1 Background

Nowadays, synthetic dyes have been widely used in many industrial processes especially in the textile industry, paper and printing, and plastics industry (Körbahti and Rauf, 2008a). Synthetic dyes are classified according to their predominant chemical structures. The structural varieties of dyes include; acidic, reactive, basic, disperse, azo, diazo, anthraquinone-based, and metal complex. These dyes have a very complex structures and low biodegradability (Bali, 2004). In addition, the highly structured polymers of these dyestuffs cause huge threat to the environment. During the production process, there was estimated around 1 – 15 % of the dyes found in the effluent (Körbahti and Rauf, 2008a). Due to its complex structures and low biodegradability, most of the dyes present in the effluent also could be carcinogenic due to their precursors and degradation products; for example azo dyes which have big percentage of synthetic dyes and degraded into carcinogenic amines (Bali *et al*., 2004).

Numerous efforts and research have been made to remove these dangerous chemical compounds. There are many traditional techniques applied in the removal process such as coagulation and flocculation, activated carbon adsorption, membrane filtration, and sedimentation. However, these methods just convert the wastewater containing dyes into secondary waste in solid form. This secondary waste has to be either treated again or dumped as such. Recent studies shown that advanced oxidation processes (AOPs), like UV/H_2O_2 (Behnajady *et al*., 2006), photocatalytic (Zhao *et al.*, 1998), Fenton and photo-Fenton processes (Çatalkaya and Şengül, 2006) and (Bali *et al*., 2004), result in promising solution towards dyestuffs detoxification and color removal. AOP based on the H_2O_2/UV system has produced high efficiency in the degradation of several types of dye that present in the industrial effluent. UV/H_2O_2 system produces ●OH radicals that become strong oxidizing agent to degrade dyes polymer into unharmed and safe substance to be discharge into the environment (Abdullah *et al*., 2007), (Bali, 2004), (Bali *et al*., 2004), and (Shu *et al.*, 2004).

The classical and conventional methods of studying the process by maintaining the other factors at unspecified constant level cannot measure the combination of parameters that affecting the experiment results. These methods also consume more time and required numerous amounts of experiments to represent the combinational effect of the parameters. With the large number of experiment, the result will be unreliable. These limitations of conventional methods can be solved by optimizing the important parameters using response surface methodology (RSM). RSM is a collection of mathematical and statistical techniques for developing, improving and optimizing processes and can be used to screen the important parameters and compute the combinational effect even with the complex interaction between parameters. RSM is to determine the optimum condition for specified parameters and to predict the future response using the response surface model. The application of statistical experimental design techniques can improved product yields, reduced process variability and experimental time; cost effective. The design also troubleshoots process problems and makes the process “robust” against external and non-controllable factors. “Robust” means relatively insensitive to these factors or influences (Montgomery, 1997). Thus the interaction between the parameters is studied and optimized using the response surface methodology.

1.2 Problem Statement

Untreated dye effluent produced by industrial process is highly colored and possesses dangerous characteristics such as high toxicity, carcinogenic in nature, low biodegradability, and reduce sunlight penetration. It also inhibits aquatic microorganism growth and threatening the flora and fauna stability. Moreover it can cause intestinal cancer and cerebral abnormalities in fetuses for mammals especially human.

Due to the high level risk by untreated dye effluent, many treatment methods and strategies have been used to degrade the dye and minimized the risk. The application of conventional methods; coagulation/flocculation, filtration, activated carbon adsorption, sedimentation, etc. does not totally degrade the dye effluent. The biological treatment is not a solution to this problematic due to the low biodegradability or toxicity of some dyes. Meanwhile, the chemical methods have not produce sufficient reduction in organic matter and not adequate enough for decoloration of dyes. Furthermore, both methods produced secondary pollutants that required further treatments.

The photocatalytic reaction is favor over other conventional and classical method for degradation of dyes because its simplicity of the system and full degradation of dyes based on the generation of highly reactive hydroxyl radicals, appear as emerging alternatives for the mineralization of organic pollutants. The system also promises a good efficiency for the degradation process. In addition, the system does not required further treatment although using chemical such as H_2O_2. This will save the operation cost for promising wastewater treatment.

Literally, Rose Bengal (C.I. name is Acid Red 94) dye has been used in numerous application in various areas such as laboratory research, biomedical, and biological application. Despite the numerous applications, information on its photolytic decolorization is not yet available in literature. Thus, it is very important to initiate the study on Acid Red 94 (AR94) since it possessed huge threat to the environment same as the other industrial dyes application.

In this study, the UV/H_2O_2 system will be used to degrade AR 94 while the optimum condition for the photodegradation of AR 94 will be determine using Response Surface Methodology (RSM) method with the aid of Design Expert® 7.1.6 software.

1.3 Objective

The objective of this research is to improve the Acid Red 94 decolorization using experimental design application.

1.4 Scopes of Study

This study will cover following scopes:

i. Using of full factorial design to screen the significance parameters for the Acid Red 94 decolorization.
ii. Application of Response Surface Methodology for optimizing the process of photodegradation of Acid Red 94.

CHAPTER 2

LITERATURE REVIEW

2.1 Dyes and Pigments

2.1.1 History

Dyes for thousand of years literally originated from vegetable or animal resources. Roots, berries, flower, insects, and crustacean cells were combined with minerals called mordants to get the desired colours that ranged based on the colour spectrum. No organic source was considered too off-beat if it produced a quality and satisfactory colours. A change in colorant history occurred in 1856, when English chemist William Henry Perkin (1838–1907) discovered a way to produce a dye in the laboratory when he tries to synthesize quinine (the only effective antimalaria treatment present) using coal tar. Although he was not successful, but he focuses his studies on the reaction of other coal tar bases including a mixture of aniline and toluidine. After that, he was able to produce the crude bases in the presence of MeOH. That dye, mauve, was produced from materials found in common coal tar. Perkin's discovery showed chemists that dyes and pigments could be produced synthetically (Zollinger, 2004). Periodically, a numerous and wide variety of colours have flooded into the world of dye and textile.

2.1.2 Dyes

Dyes are an important class of chemicals which are widely used in many industrial processes such as the leather, textile, and printing industries (Rauf *et al*., 2007). There are various kinds of dyes available in market such as azo, anthraquinone, triarylmethane, diarylmethane, acridine, quinine, xanthenes, and nitro dyes. These dyes are extensively used to impart colour to various industrial applications (Rauf *et al*., 2008). Shigwedha *et al*., (2007) found that textile industries are the largest consumers of organic dyes and estimated around 10 – 15 % of the dyes used are lost during the dyeing process and released into the effluents.

Synthetic dyes are an important class of chemicals which are used in many industrial processes. Synthetic dyestuffs have complex chemical structures which is not easy to degrade biologically. Evenmore, many industrial many biodegradability studies on dyes have showed that they are not likely biodegradable (Bali, 2004). Most of the dyestuffs are highly structured polymers with low biodegradability (Rauf *et al*., 2008). Dyes also usually contain elements such as nitrogen, chlorine, and sulphur. The oxidation products of these elements may be higher in toxicity than the parent molecule (Chen *et al.*, 2005). Particularly, synthetic dyes contribute to special environmental concern due to their degradation products such as aromatic amines which are considered as highly carcinogenic substances (Bali *et al*., 2004).

The wastewater containing dyes have a great variety of organic contaminants in a wide range of concentration especially in textile industry where they are highly coloured and complex variable of nature (Kurbus *et al*., 2003). The disposal of these coloured wastewaters poses huge problem for industry as well as a threat to the environment (Behnajady *et al*., 2006). It is because many of dyes are hard to be removed as they are stable to light and heat and are biologically non-degradable (Chen *et al*., 2005). In addition, the coloured dye effluents are considered to be highly toxic to aquatic life and and affect the symbiotic process by disturbing the natural equilibrium and reducing photosynthetic activity and primary production due to the colorization of water.

Effluents also contain significant level of organic contaminants which is toxic as they create odor, bad taste, unsightly colour and foaming (Ravikumar *et al*., 2006). Thus decoloration of effluents from dyes industrial application was happened to be a very important because of aesthetic and environmental concerns (Attia *et al*., 2008).

2.1.3 Rose Bengal (Acid Red 94)

Acid Red 94 (AR 94) is a tetraiodo-substituted dye of the xanthene class of dyes. It exhibits unusual spectroscopic and photochemical properties including a huge absorption coefficient in the visible region and a high tendency for intersystem crossing to produce a photochemically active triplet excited state. Figure 2.1 shows the molecular structure of Acid Red 94.

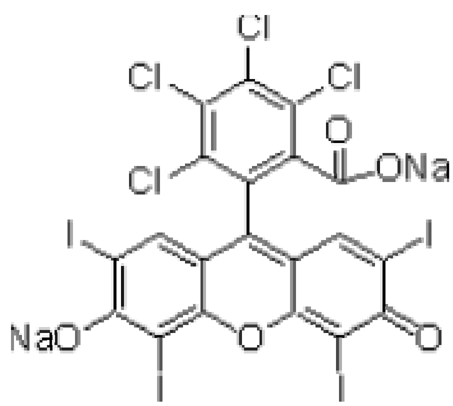

Figure 2.1: Molecular structure of Acid Red 94

The dye has been applied in photodynamic inactivation of catalase (Kim *et al.*, 2001), photoinactivation of $NADP^+$ via the production of singlet oxygen (Kim *et al.*, 2004), as a photosensitizing agent for inactivating biological species such as vaccinia virus, microsomal glucose-6-phosphatase (Lenard *et al.*, 1993), tripsin, *Escherichia coli* (Kita *et al.*, 1984), acetylcholinesterase, and HL-60 cells (Schäfer *et al.*, 2000).

2.2 Degradation of Dyes

2.2.1 Conventional Methods

Many efforts have been devoted to develop technologies that are able to minimize the hazardous effects caused by dye based industrial activities. The many different conventional methods applied in industrial wastewaters, such as coagulation and flocculation, membrane separation (ultra filtration, reverse osmosis) or elimination by activated carbon adsorption are not sufficient enough. This is because these process likely to produce a secondary pollutant or dumped as such (Körbahti and Rauf, 2008b). Then, this secondary pollutant is either sorbed or trapped in bioflocs. Thus, ecosystems of streams can be seriously affected (Bali, 2004). The United States Environmental Protection Agency's (U.S. EPA) Water Engineering Research Laboratory first reported that 11 out of 18 studied azo dyes (synthetic dyes) were substantially unaffected by the activated sludge process. Generally, adsorption onto activated carbon or chemical coagulation was applied to deal with wastewater containing dyes. However, these treatments also mainly transferred the contaminant from wastewater into solid wastes that the spent activated carbon and coagulant sludge need further and ultimate disposal eventually (Shu *et al*., 2004).

Removal of dyes from effluents in an economic way remains a huge problem for textile industries. Adsorption technique is an excellent way to treat effluents more than other conventional process, especially from the environmental point of view (Ravikumar *et al*., 2006). Carbon is being used as an adsorbent because of its high efficiency in treating the organic materials in effluents. Although it possessed high efficiency, but the enhancement of the price of activated carbon result in increasing the cost for its operation (Khattri and Singh, 1998). Biological treatment of wastewater can eliminate the important organic compounds. However, the biochemical decomposition by conventional method does not adequate enough to completely decolorize dye effluents

(Çatalkaya and Şengül, 2006). Thus it is a necessity for dyes based industry to reconsider upon the alternative method in their wastewater treatment.

2.2.2 Advanced Oxidation Processes

Advanced oxidation processes (AOPs) are alternative methods for decolorizing and reducing wastewater effluents generated by industries (Abdullah *et al*., 2008). AOPs are effective for detoxification and mineralization of the effluents from textile dyeing mills (Szpyrkowicz *et al*., 2001). Behnajady *et al*., (2006) stated that AOPs also a non-destructive physical water treatment processes, because they are able to eliminate compound rather than changing them into another medium such as solid waste. The use of AOPs, like UV/H_2O_2 (Körbahti and Rauf, 2008a), photocatalytic (Attia *et al*., 2008), Fenton and photo-Fenton processes (Çatalkaya and Şengül, 2006), has shown promising results as these processes appear to have the ability to completely decolorize and partially mineralize the textile industry dyes in short reaction time (Rauf *et al*., 2008), (Körbahti and Rauf, 2008b) and (Bali *et al*., 2004).

Among the AOPs, chemical oxidation using UV in the presence of H_2O_2 is a very promising technique. Process involving the use of UV radiation and H_2O_2 are characterized by the generation of hydroxyl radicals (Behnajady *et al*., 2006). UV wavelengths of 200 – 280 nm lead to dissociation of H_2O_2, with mercury lamps emitting at 254 nm being the most used. UV/H_2O_2 systems generate hydroxyl radicals (●OH) which are highly powerful oxidizing agents. Hydroxyl radicals can oxidized organic compounds (RH) producing organic radicals (●R), which also highly reactive and can be further oxidized (Bali *et al*., 2004). These radicals can then attack the dye molecules to undergo a series of reactions in which the organic molecules will be eliminated or converted into a simple molecules or harmless compound (Abdullah *et al*., 2007). The main reaction that occurs during UV/H_2O_2 oxidation process is as follows:

$$H_2O_2 + UV \rightarrow 2OH\bullet \qquad (2.1)$$

$$H_2O_2 \leftrightarrow HO_2^- + H^+ \qquad (2.2)$$

$$RH + OH\bullet \rightarrow H_2O + R\bullet \rightarrow \text{further oxidation} \qquad (2.3)$$

where R is the carbon chain.

The hydroxyl radicals will oxidize organic compounds producing organic radicals, which also a highly reactive and can undergo further oxidation. When generated, these radicals will react quickly and usually randomly with most organic compounds. The resulting organic radicals then reacts with oxygen to initiate series of degradative oxidation reaction that lead to mineralization of products such as CO_2 and H_2O (Çatalkaya and Şengül, 2006). The other possible reactions that may occur during the UV/H_2O_2 process are hydrogen abstraction, electrophilic addition and electron transfer reactions (Behnajady *et al.*, 2006). Although AOPs have much kind of advantages in dyes decoloration, one major problem in AOPs is the high energy demand for UV lamps which lead to high operational cost. In order to minimize the irradiation time, energy consumption, and operational cost, there is necessary to optimize the pH condition, chemical types, chemical concentration, and pollutant/oxidant ratio, therefore are very important (Çatalkaya and Şengül, 2006). For this aim, the application of experimental design is the best solution where it will be used to optimize the important parameters that affected the efficiency of dyes decoloration.

2.3 Experimental Design Application

2.3.1 Introduction

Experimental design is a very important application in chemometrics (application of mathematical or statistical methods to chemical data), because chemical experiments have to be performed to get more knowledge about a process or system. The science is dependent on the experiments while the experimental design is to improve the experimental works. Experimental design methodology is used to decide which experiment needs to take place in order to get information on the certain chemical processes or products. It is used to determine which factors have an influence on the process output. Another application is to decide on how many experiment need to take place in order to obtain adequate knowledge on the desired product and the system. Thus, it is clear that optimization is an essential in chemical or biochemical processes (Kurbus *et al*., 2003).

In the other hand, the main objective of the experimental design (DoE) is to determine, with a minimum effort (less number of experiments), the effects of the different factors and their interactions in the process response, within the range of the studied variables. Besides, the information obtained allows deciding which factors and/or interactions are statistically significant. For that purpose, statistical techniques need to be used such as analysis of variance (ANOVA).

2.3.2 Screening

According to Kurbus *et al*., (2003), main methods of experimental design are factorial design including full factorial, fractional factorial design, orthogonal design (OD), D-optimal design, and uniform design. The selection of experiments has particular influence on the system. It is applied to determine the conditions to get the product of a process with desirable characteristics. The characteristics of the product named as a response. The factors that affecting the product are called independent variables while the product or the response is called dependent variables. So, experimental design is a set of carefully planned experiments.

The main step in experimental design is to choose the initial factors and response and to select the experimental domain. After screening process, unimportant factors are discarded and type of experimental design is determined beforehand. Normally, two level factorial designs are used for determination of significance parameters and intervals. The optimal response usually the lowest or the highest value of process output or response. After determining which factors have minimal or insignificance on the response, the optimum settings of the significant parameters levels that produce the best response need to be performed.

2.3.3 Optimization

According to Çatalkaya and Şengül, (2006), in order to find the optimum reaction conditions and to study the effect of significance parameters of dyes decoloration, the response surface methodology (RSM) was used. RSM essentially is the set of mathematical and statistical methods for designing experiments, building models, evaluating the effect of variables, and finding the optimum conditions of variables to predict the target responses (Myers and Montgomery, 2002). It is an important branch of experimental design and a critical tool in developing new processes, optimizing their performance, and improving design and formulations of new products (Körbahti and Rauf, 2008a). RSM is used for the analysis of dependent variables as functions of independent variables. Response surface procedures are not only primarily used for the purpose of allowing the researchers in order to understand the mechanism of the system or process but the most importantly is to determine the optimum operating conditions or to determine a region for the factors at a certain operating specification (Rauf *et al*., 2008).

2.3.4 Application of Experimental Design in Photodegradation of Dye

Experimental design greatest applications have been in industrial research, specifically in situations where there are large numbers of variables influence the system feature. This feature termed as response and normally measured on continuous scale, represents the most important functions of the systems (Myers and Montgomery, 2002).

Although initially, RSM was developed for the purpose of the chemical industry application, but now it has been used in various field and wide applications in physical sciences and engineering as well as in biological, clinical, and social sciences (Khuri, 2001). In this study, response surface design was selected as it provides a reasonable distribution of data points throughout the areas of interest, allows model adequacy including lack of fit, allows design of higher order to be built up sequentially and provides internal estimate of error. In addition, the response surface designs do not require a large number of runs and also do not require too many levels of independent factors (Montgomery, 1996).

Another part of this study also was to use RSM to find a suitable approximating function in order to predict and determine the future response as well as to investigate the optimum operating conditions in the areas for which the factors at a certain operating specifications are met.

Meanwhile, there was numerous study carried out on dye degradation using experimental design application. Bali (2004) and Çatalkaya and Şengül, (2006) were employed the Box-Wilson experimental design to evaluate the effects of major process variables on degradation efficiency. Bali (2004) was used three azo dyes in his study (Direct Red 28, Direct Yellow 12, and Reactive Black 5) while Çatalkaya and Şengül, (2006) used bakery's yeast industry effluent and both experimental result turned out to be followed the first-order reaction law.

Körbahti and Rauf (2008a) optimized the photolytic degradation of Basic Red 2 (BR 2) using response surface methodology (RSM). The factors studied were dye and H_2O_2 concentration and pH condition. They have found that RSM is a powerful tool in developing the equation model and successfully optimizing the conditions of photo-oxidation process of dye. Furthermore, the design of experiment application was also help graphically and statistically to analyze the experimental data.

CHAPTER 3

METHODOLOGY

3.1 Introduction

The best available technique for degradation or decoloration of dyes is the advanced oxidation processes (AOPs). One of the effective processes is the photolytic reaction using UV/H_2O_2 system. This method is used since the system has high degree of degradation efficiencies yet the simplest system among AOPs. The main objective of the study is to improve the efficiency of dye decoloration using statistical design analysis. Thus, the important parameters will be optimized. Upon the optimization, the response surface methodology will be applied exploiting central composite design (CCD) analysis. Finally, the optimization will produce a model equation that useful in predicting the future response using the same parameters.

3.2 Samples Preparation

The samples were prepared based on Rauf *et al.*, 2008. Acid Red 94 stock solution of 0.001M was prepared in 100mL of deionized water in a 250mL flask. Desired concentrations of the samples are done by dilutions of the stock solution using deionized water. The series of the diluted samples were mixed with a given amount of H_2O_2 and the mixture was irradiated with UV light for different periods of time. After certain time of irradiation, the absorbance of the solution was monitored instantaneously in UV-Vis Spectrophotometer. The percentage decoloration of the dye solution was obtained by monitoring the change in absorbance value of each case. The temperature of this experiment was maintained at 25 ± 2 ^{o}C.

Percentage of decoloration was calculated using following equation:

$$\textbf{Percent decolorization} = (A_i - A_f)/A_i \qquad (3.1)$$

where A_i is the initial absorbance and A_f is the final absorbance value.

3.3 Experimental Design Application

3.3.1 Screening Process

In this study, the important parameters were screened using two-level factorial design with full factorial analysis. The purpose of the screening process was to identify the significant parameters of the decolorization process.

The matrix design of the experiment was developed based on the design in the Design Expert Software® 7.1.6. A 2^4 full factorial design was used to show the statistical significance of the medium composition. The factors were Acid Red 94 (AR 94) concentration, H_2O_2 concentration, pH and time. A total of 16 experiments were employed to determine the significant factors affecting the percent of AR 94 degradation. The settings of range for factors were based on the investigation of single factors (screening process) and literature. The experiment results were then analyzed using the software and insignificant parameter was screened out.

Table 3.1: Factors (variables) studied and their concentration levels.

Variables	Unit	Actual Value		Coded Value	
		Low	**High**	**Low**	**High**
AR 94 concentration	µM	20	160	-1	1
H_2O_2 concentration	M	0.01	0.05	-1	1
pH	-	3.75	10.00	-1	1
time	min.	0	30	-1	1

Coding of the variables was done according to the following equation:

$$x_i = \frac{(X_i - X_0)}{\Delta X_i} \quad (3.2)$$

where, x_i = coded value of the *i*th independent variable,

X_i = actual value of the *i*th independent variable,

X_0 = actual value of the *i*th independent variable at the centre point,

ΔX_i = step change.

The coded values for different variables are shown in Table 3.1 and the design matrix of the full-factorial design is shown in the Table 3.2. The positive and negative signs refer to levels -1 and +1 respectively, of the experimental plan at two levels as -1 and +1 indicate minimum and maximum value for each coded variable in the studied factors. Finally, among the 16 runs to perform (Table 3.2) all runs correspond to factorial design.

Table 3.2: The matrix of two-level full factorial design.

Run	Factor 1 (x_1)	Factor 2 (x_2)	Factor 3 (x_3)	Factor 4 (x_4)
	A: [AR 94]	B: [H_2O_2]	C: pH	D: time
1	-1	-1	-1	-1
2	1	-1	-1	-1
3	-1	1	-1	-1
4	1	1	-1	-1
5	-1	-1	1	-1
6	1	-1	1	-1
7	-1	1	1	-1
8	1	1	1	-1
9	-1	-1	-1	1
10	1	-1	-1	1
11	-1	1	-1	1
12	1	1	-1	1
13	-1	-1	1	1
14	1	-1	1	1
15	-1	1	1	1
16	1	1	1	1

3.3.2 Optimization Process

The optimization process was done after the screening process. The decoloration of dyes was optimized by response surface methodology (RSM). The runs were designed with central composite design (CCD) using Design Expert® 7.1.6 software. The CCD was randomized the parameters interaction and proposed the optimum condition with the combinational effect of the parameters. After obtaining the experiment layout, the experiment was run accordingly. The results data were analyzed using Design Expert @ 7.1.6 as well to find the optimum condition with highest percentage of dyes decoloration.

Table 3.3: The design matrix of central composite design

Std	Factor 1 A:Dye uM		Factor 2 B: H_2O_2 M		Factor 3 C:pH		Factor 4 D:time min	
1	-1	20	-1	0.01	-1	3.75	-1	0
2	1	160	-1	0.01	-1	3.75	-1	0
3	-1	20	1	0.05	-1	3.75	-1	0
4	1	160	1	0.05	-1	3.75	-1	0
5	-1	20	-1	0.01	1	10	-1	0
6	1	160	-1	0.01	1	10	-1	0
7	-1	20	1	0.05	1	10	-1	0
8	1	160	1	0.05	1	10	-1	0
9	-1	20	-1	0.01	-1	3.75	1	30
10	1	160	-1	0.01	-1	3.75	1	30
11	-1	20	1	0.05	-1	3.75	1	30
12	1	160	1	0.05	-1	3.75	1	30
13	-1	20	-1	0.01	1	10	1	30
14	1	160	-1	0.01	1	10	1	30
15	-1	20	1	0.05	1	10	1	30
16	1	160	1	0.05	1	10	1	30
17	-α	20	0	0.03	0	6.875	0	15
18	α	160	0	0.03	0	6.875	0	15
19	0	90	α	0.01	0	6.875	0	15
20	0	90	α	0.05	0	6.875	0	15
21	0	90	0	0.03	α	3.75	0	15
22	0	90	0	0.03	α	10	0	15
23	0	90	0	0.03	0	6.875	α	0
24	0	90	0	0.03	0	6.875	α	30
25	0	90	0	0.03	0	6.875	0	15
26	0	90	0	0.03	0	6.875	0	15

The experimental results obtained from above design matrix for the desired response, Y_i, are then fitted to a quadratic model (Eq. 3.3) that takes in account the linear and quadratic effects of the n variables, as well as the interaction among them. The least square fitting was performed using Design-Expert® 7.1.6 software as well.

$$Y_i = \beta_o + \sum_{i=1}^{n} \beta_i x_i + \sum_{i=1}^{n} \beta_{ii} x_i^2 + \sum_{j>i}^{n} . \sum_{i=1}^{n} \beta_{ij} x_i x_j \qquad (3.3)$$

In equation 3.3, x_i refers to coded variable associated to parameter i (in this study it varies between 1 to 4; AR 94 concentration, H_2O_2 concentration, pH, and time); β_o is the interception term and corresponds to the response value when x_i is null for all variables; β_i determines the influence of parameter i in the response (linear term); β_{ii} is a parameter that determine the shape of the curve (quadratic effect); and finally β_{ij} reflects the effects of the interaction among variables i and j. The least-square regression analysis provided the estimates of these coefficients in the second-order model.

The statistical analysis then proceeds with an analysis of variance (ANOVA), which evaluates the adequacy of the model fitting.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Screening the Optimum Decolorization Conditions by Full Factorial Design (2^4 Design)

Screening studies on the factors of dye decolorization conditions were carried out by using 2^4 full factorial design. This study was done to determine the significant factors for dye decolorization. The factors descriptions were obtained from Rauf *et al*. (2008). The factorial experiment carried out in a batch mode experiment to study the factors expected to influence the percent of Acid Red 94 (AR 94) decolorization. The four factors are AR 94 concentration (A), H_2O_2 concentration (B), pH (C), and time (D).

Each factor was presented in low and high level, and the data obtained from a single replicate of the 2^4 experiment shown in Table 4.1. All the 16 runs were made in random order by Design Expert® 7.1.6 software.

Table 4.1: Full factorial design matrixes with experimental value of percent degradation

Std	Run	Block	Factor 1 A:Dye uM	Factor 2 B: H_2O_2 M	Factor 3 C:pH	Factor 4 D:time min	Response 1 % degradation
1	13	Block 1	20	0.01	3.75	0	53.17
2	12	Block 1	160	0.01	3.75	0	89.74
3	14	Block 1	20	0.05	3.75	0	89.82
4	3	Block 1	160	0.05	3.75	0	75.36
5	16	Block 1	20	0.01	10	0	14.93
6	11	Block 1	160	0.01	10	0	10.85
7	8	Block 1	20	0.05	10	0	3.85
8	2	Block 1	160	0.05	10	0	0
9	4	Block 1	20	0.01	3.75	30	56.79
10	5	Block 1	160	0.01	3.75	30	91.54
11	6	Block 1	20	0.05	3.75	30	92.31
12	10	Block 1	160	0.05	3.75	30	77.99
13	9	Block 1	20	0.01	10	30	19.68
14	15	Block 1	160	0.01	10	30	12.42
15	1	Block 1	20	0.05	10	30	6.56
16	7	Block 1	160	0.05	10	30	1.18

The highest percent of AR 94 decolorization was observed at the 11th run of experiment. Mostly, high percent of AR 94 decolorization was recorded at pH 3.75 while low percent of AR 94 decolorization recorded at pH 10. Other factors did not give much influence towards AR 94 decolorization.

Normal probability plots of effects were used to assess the importance of the effects. Although the full normal probability plots is less sensitive for selecting small effects, but it could differentiate between positive and negative effects.

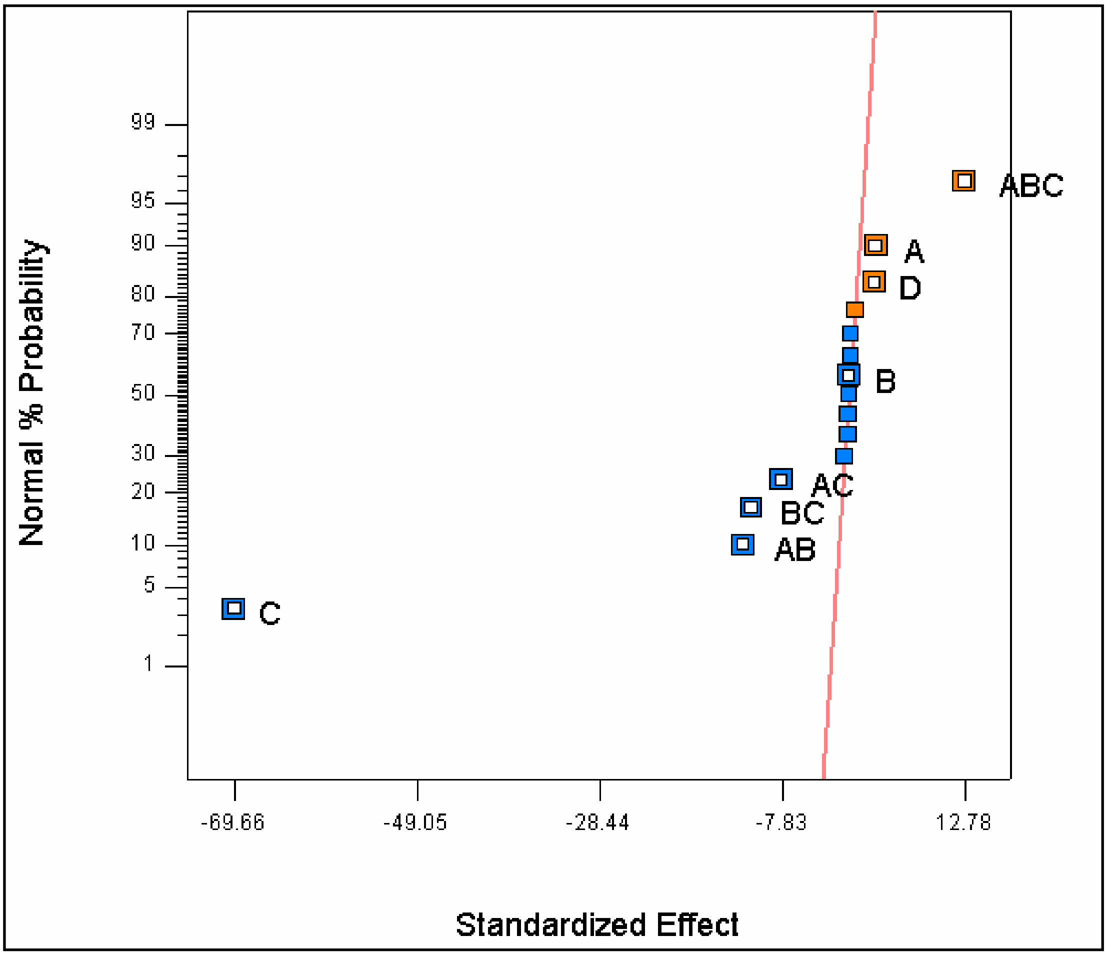

Figure 4.1: Normal probability plot of effects for AR 94 degradation

The normal probability plot of these effects is shown in Figure 4.1. All of the effects that lie along the line are negligible, whereas the large effects or the significance effect are far from the line. The important effects that emerged from this analysis were the main effects of A, C, and D and AB, AC, BC, and ABC interactions.

In using the experimental design approach, the runs were conducted in full factorial designed experiments to visualize the effects of independent factors on the response and the results along with the experimental conditions. The experimental results were evaluated and approximating function of AR 94 degradation percent obtained in Equation 4.1.

$$\hat{y} = 1.37x_1 - 0.13x_2 - 34.83x_3 + 1.30x_4 - 6.12x_1x_2 - 3.94x_1x_3 - 5.66x_2x_3 + 6.39x_1x_2x_3 \quad (4.1)$$

In Equation 4.1, $\hat{y}$ is the AR 94 degradation percent whereas x_1, x_2, x_3, and x_4 correspond to independent variables of AR 94 concentration (μM), H_2O_2 concentration (M), pH, and time (minutes), respectively.

Table 4.2: ANOVA results of the linear model of photodegradation of Acid Red 94 (AR 94) dye with H_2O_2

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F
Model	21479.5303	8	2684.9412	3996.7281	< 0.0001
A-Dye	30.1675	1	30.1675	44.9065	0.0003
B- H_2O_2	0.2626	1	0.2626	0.3909	0.5516
C-pH	19407.9726	1	19407.9726	28890.1625	< 0.0001
D-time	26.9101	1	26.9101	40.0577	0.0004
AB	600.1275	1	600.1275	893.3329	< 0.0001
AC	248.9295	1	248.9295	370.5494	< 0.0001
BC	512.2300	1	512.2300	762.4912	< 0.0001
ABC	652.9302	1	652.9302	971.9336	< 0.0001
Residual	4.7024	7	0.6717		
Cor Total	21484.2328	15			

$R^2 = 0.9998$, adj. $R^2 = 0.9995$, adequate precision = 151.401

Table 4.2 summarizes the analysis of variance (ANOVA) for this design. ANOVA results of this model show that it can be used to navigate the design space. The model F-value of 3996.73 implies the model is significant for AR.94 degradation and there’s only a 0.01% chance that a model with F-value this large could occur due to noise. In AR 94 degradation model, the adequate precision ratio of 151.401 indicates an adequate signal where it measures the signal to noise ratio; a ratio greater than four is desirable. The p-values less than 0.0500 indicate that the model term are significant, whereas the values greater than 0.1000 are usually considered significant. However, it is more dependable and easier to use p-value to determine significant factor for the response model. Thus, the model and the terms are significant according to p-value except for H_2O_2 concentration factor where the p-value is 0.5516. However, it is unnecessary to exclude the factor as it does not affect model adequacy and adjusted R^2 very much.

Generally, the effects of experimental factors on degradation percentage response are based on Equation 4.1 which generated from the response data and analysis of variance. One of the important parts of data analysis procedure is the model adequacy check as the approximating model would measure the poor or misleading results if it were an inadequate fit model. This is done by analyzing the residual plots which are examined for the approximating model (Myers and Montgomery, 2002). The normal probability and studentized residuals plot is shown in Figure 4.2.

The normal probability plot of studentized residuals gave an illustration on the normality assumption. The assumption was satisfied if and only if the residuals plot approximately along the straight line.

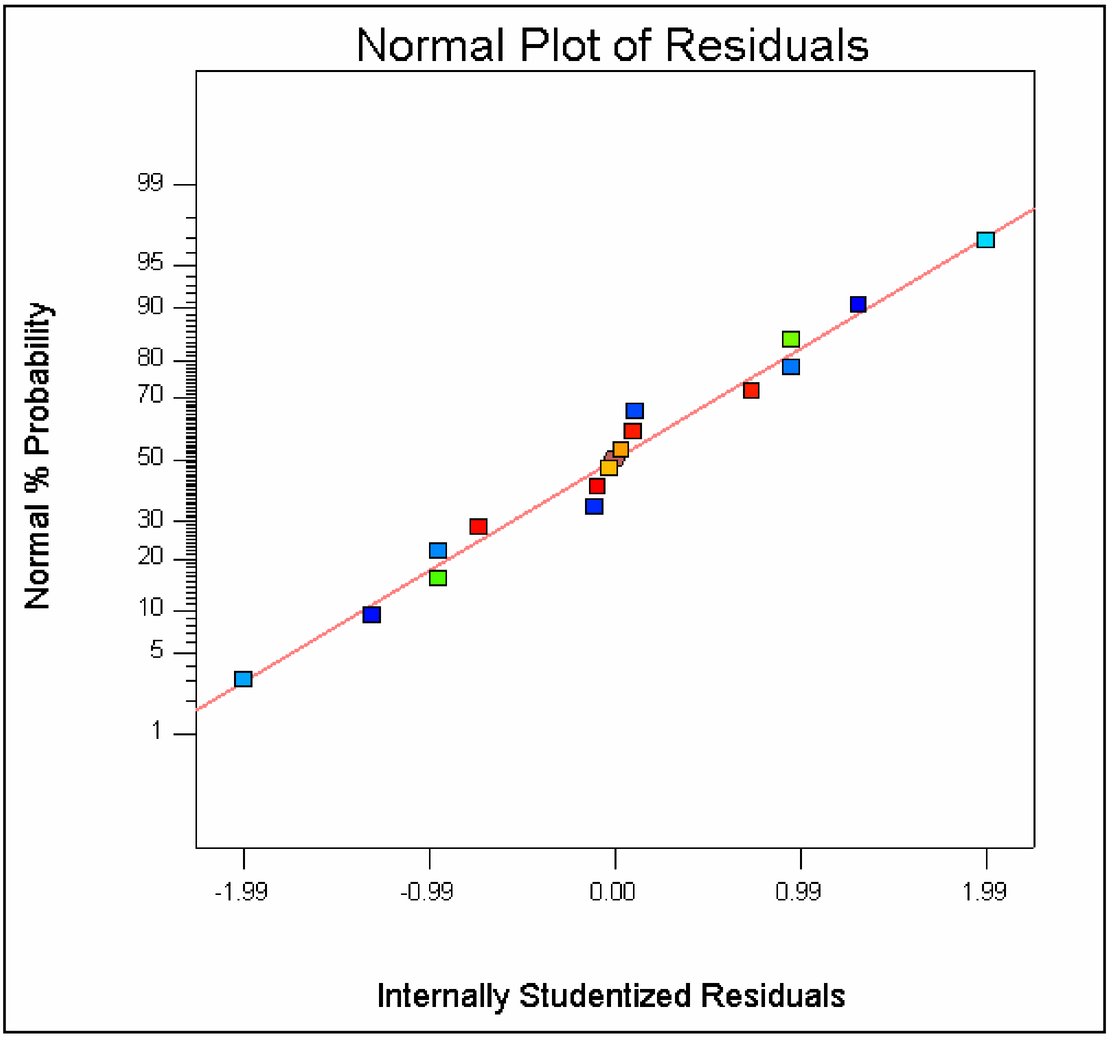

Figure 4.2: The studentized residuals and normal percentage probability plot of photodegradation of Acid Red 94.

In Figure 4.2, residuals show how well the model satisfies the assumptions of analysis of variance (ANOVA) whereas the studentized residuals measure the number of standard deviations separating the actual and predicted values. For overall observation, Figure 4.2 shows that neither response transformation was needed nor there was any obvious deviation from the normality line. Thus, the model's normality assumptions are satisfied.

Figure 4.3 shows the studentized residuals versus predicted AR 94 percent degradation. The general characteristic is that the data should be randomly scattered, suggesting the variance of original observations is constant for all values of the response. If the variance of the response depends on the mean level of y, then this plot often exhibits a funnel-shaped pattern (Myers and Montgomery, 2002). However, Figure 4.3 shows that the result data were randomly scattered inside the boundary which means there was no need for transformation of the response variable.

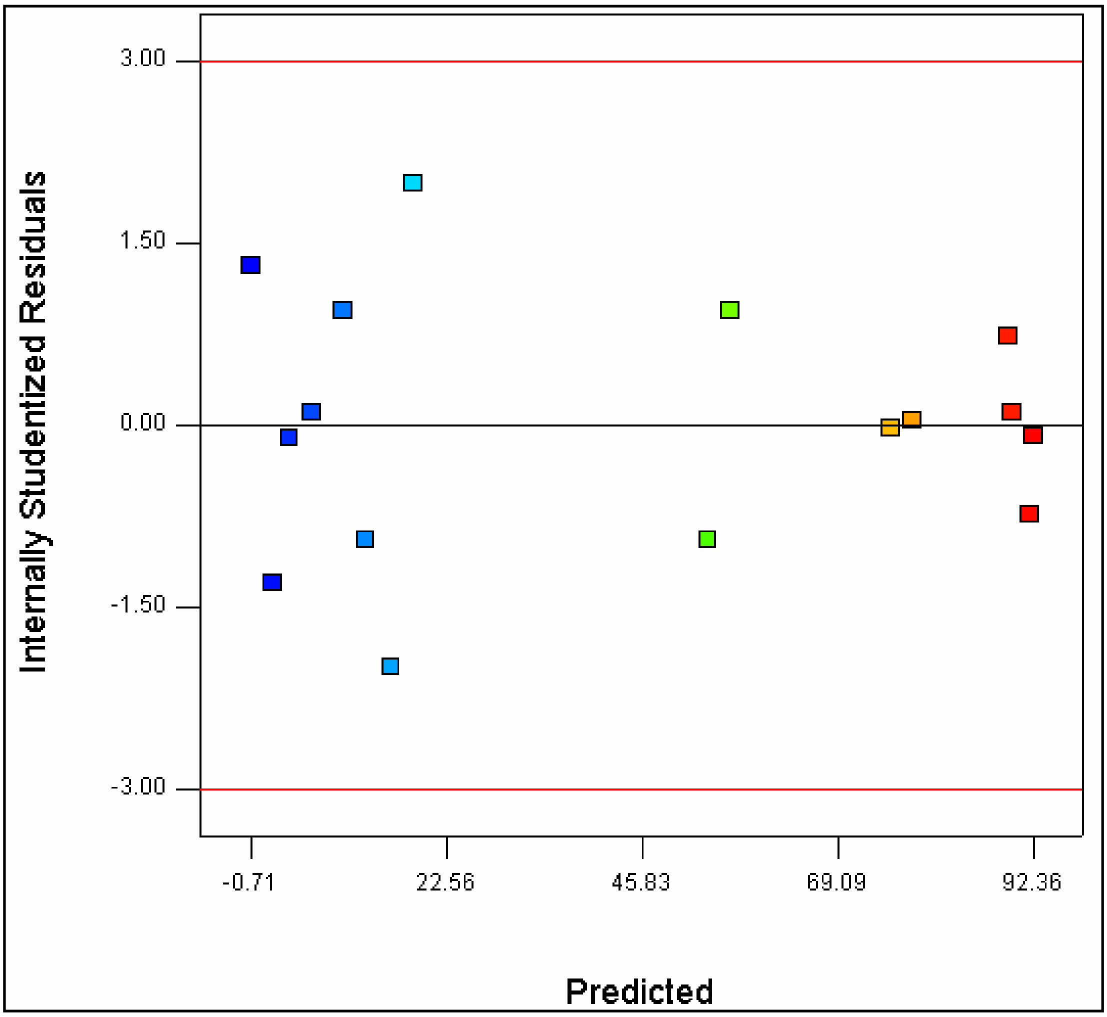

Figure 4.3: The predicted degradation of Acid Red 94 dye and studentized residuals plot.

The actual and predicted AR 94 percentage degradation are shown in Figure 4.4. Actual values were the measured response data for a particular run, and the predicted values were evaluated from the model and generated by using the approximating functions. The values of R^2 and adjusted R^2 were found to be 0.9998 and 0.9995 respectively. Thus, the size of the studentized residuals are said to be independent of its predicted values. In other words, the spread of the studentized residuals approximately the same across all levels of the predicted values which means the plot was fine.

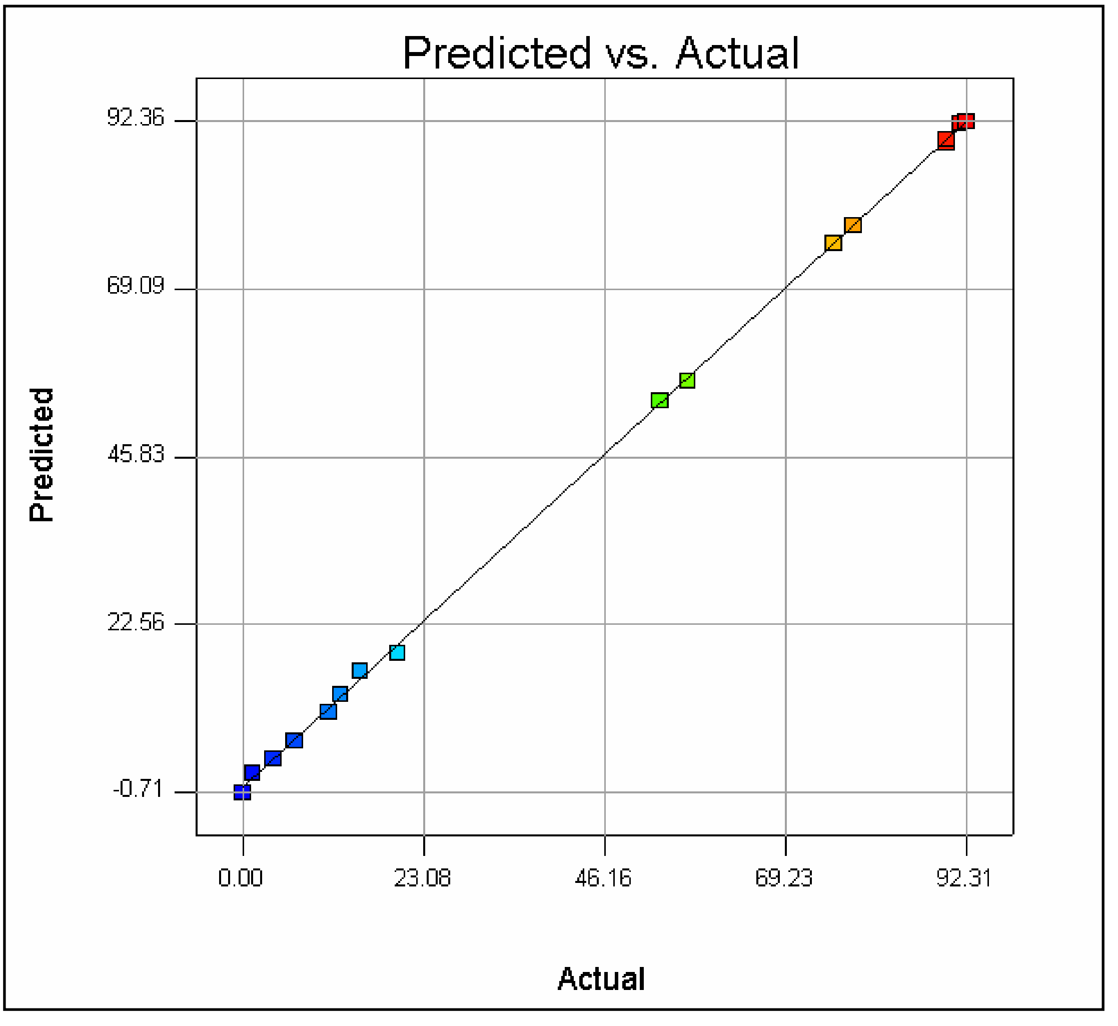

Figure 4.4: The actual and predicted plot of Acid Red 94 degradation (R^2 = 0.9998, adj. R^2 = 0.9995).

4.2.1 Effect of Acid Red 94 Concentration and H_2O_2 concentration on Percent of Dye Degradation using One Factor Plot

Effect of AR 94 concentration on the percentage of dye degradation is shown in Figure 4.5. The slope was quite plain and almost horizontal as percent degradation increased slightly with increasing in dye concentration from 20µM to 160µM. This means dye concentration effect was not the significant factor for the degradation process. For H_2O_2 concentration factor, it was clearly proved in the normal probability plot that it was an insignificant factor for percent degradation response.

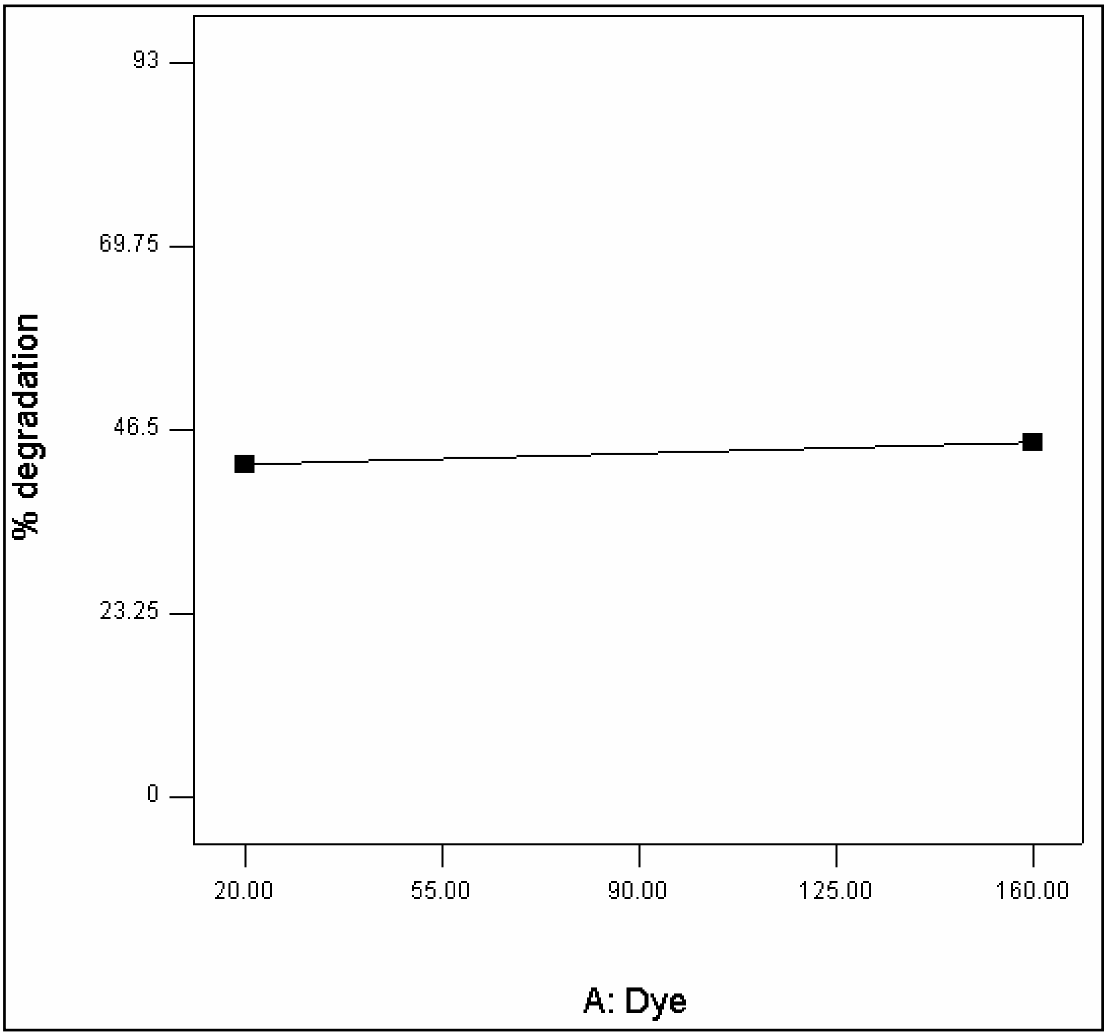

Figure 4.5: Effect of dye concentration on percent of dye degradation.

In other research conducted by Rauf *et al*. (2008), dye concentration factor shows a significant effect on percent of dye degradation. Generally dye decolorization increased with decreasing in dye concentration. Low dye concentration will result in higher percentage of AR 94 decolorization.

Similar trend was also observed for H_2O_2 concentration effect. The difference in H_2O_2 concentration does not give a significant effect on the percent of AR 94 decolorization (Figure 4.6).

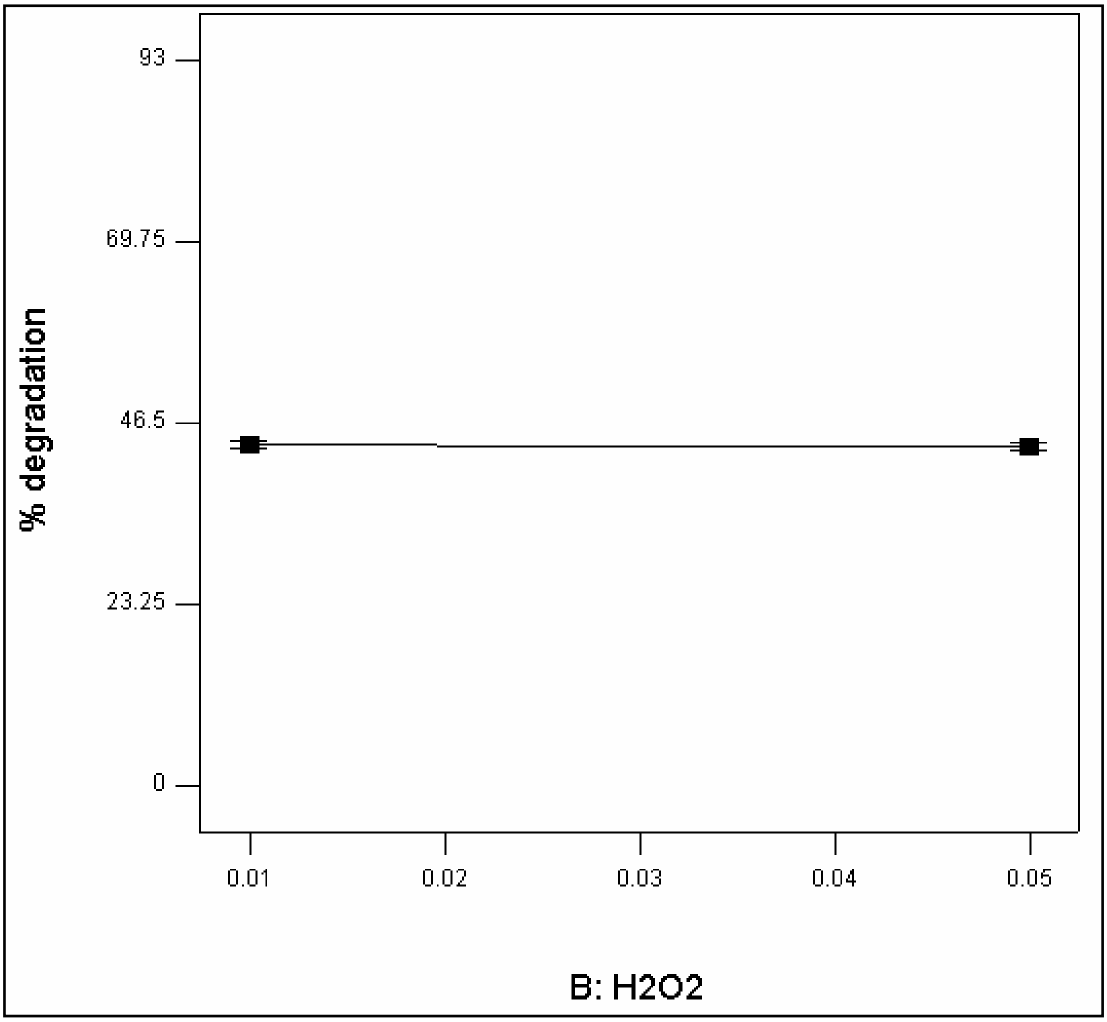

Figure 4.6: Effect of H_2O_2 concentration on percent of dye degradation.

Again, there was a contradiction with the scientific facts that the decolorization of dye solution is due to the reaction of hydroxyl radicals generated by hydrogen peroxide in solution upon UV light irradiation (Aleboyeh et al., 2005). In the other words, higher concentration of H_2O_2 resulted in high percent of dye degradation.

4.2.2 Effect of pH and time on Percent of Dye Degradation using One Factor Plot

Figure 4.7 shows the effect of pH on dye degradation response. The desired response was tested at pH 3.75 and 10.00 with maximum percent of dye degradation have been observed at low pH value and decrease significantly along with the increase in pH value.

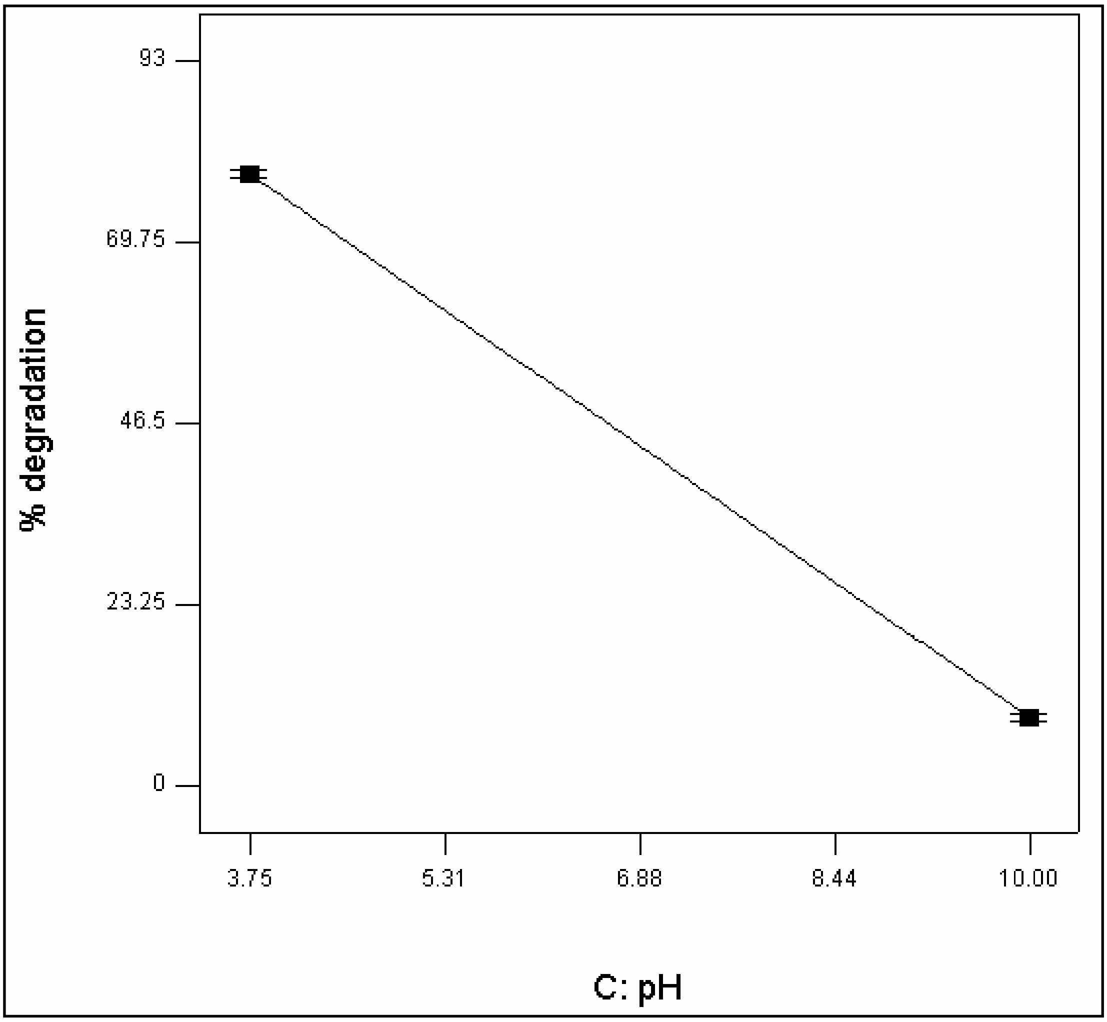

Figure 4.7: Effect of pH on percent of dye degradation.

However, Rauf *et al*. (2008) found that although pH have significant effect, but the optimum condition which give the highest percent of degradation was observed at pH 6.6. The enhancement in dye decolorization in less acidic conditions is most likely due to the fact that peroxide anions (HO_2^-) are produced in solution by UV radiation, which in turn generated more radicals (Aleboyeh *et al*., 2005).

On the other hand, time does not have any significant effect on the desired response since the dye degradation slightly increases through the time factor as shown by Figure 4.8.

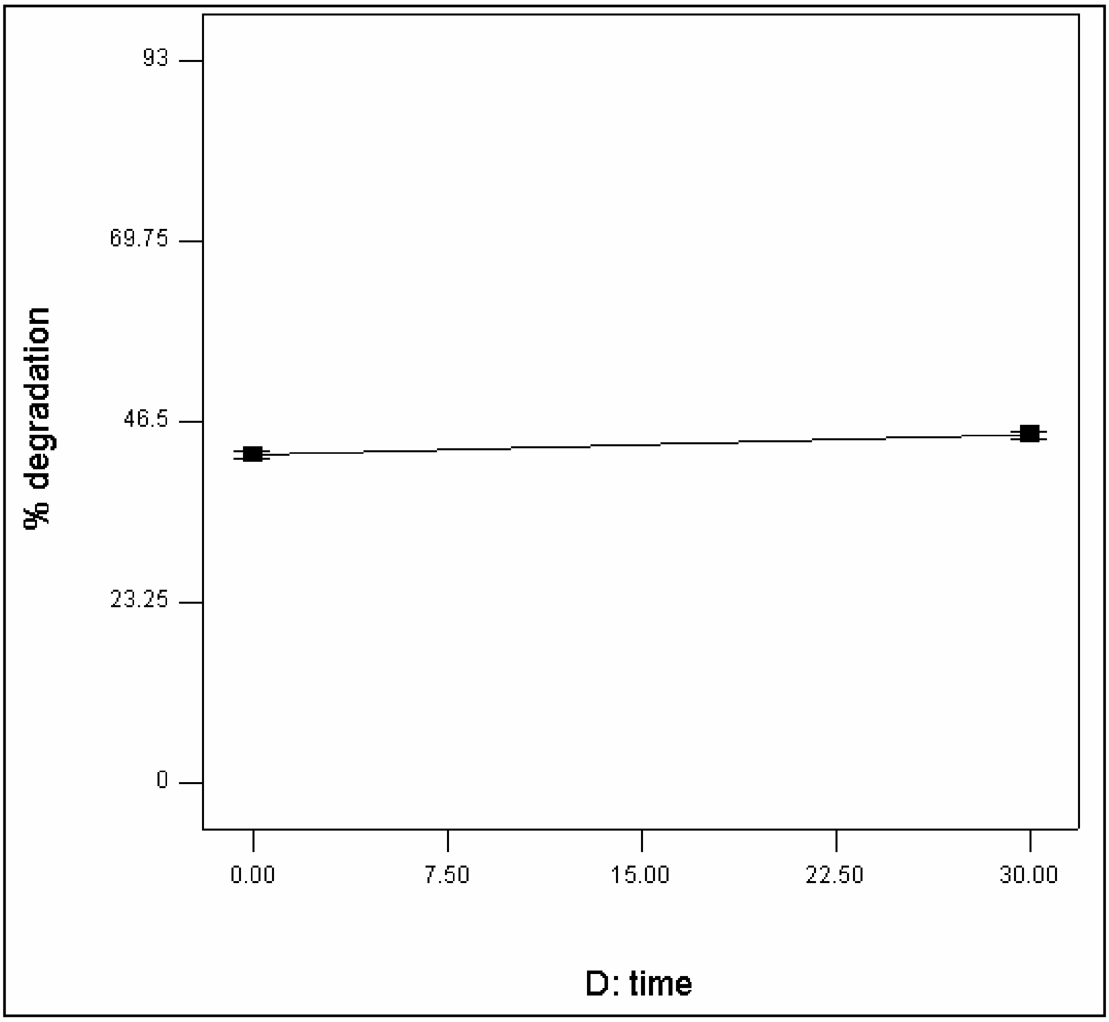

Figure 4.8: Effect of time on percent of dye degradation.

The time effect study by Körbahti and Rauf (2008b) also resulted in insignificant effect of time. This is because, dye decolorization reaction was solely depend on the hydroxyl radicals that present in the dye solution not how long the irradiation time take place.

Based on all the parameter's effects, it can be concluded that pH was the only significant factor for AR 94 decolorization process. Eventhough the scientific facts do not supported the result, but the experiments were conducted based on the proposed conditions by approved literature. The opposite results were likely due to unfavourable experimental conditions which then lead to significant noise factors effect.

4.3 Analysis by Response Surface Methodology (RSM)

In the first step of the study, the effect of operating parameters of initial dye concentration, H_2O_2 concentration, pH, and time on decolorization of Acid Red 94 (AR 94) dye was investigated using full factorial design. In the second step, the objective was to select the optimal point of decolorization of Acid Red 94 (AR 94) dye. The result data from the full factorial design was then augmented to be conducted in central composite design experiments to visualize the effects of independent factors on the response and the results along with the experimental conditions (Myers and Montgomery, 2002). By using central composite design (CCD), the experiments different combination of AR 94 concentration, H_2O_2 concentration, pH, and time will be performed. Experiments arranged by Design Expert@ 7.1.6 software are listed in Table 4.3 along with the actual and predicted response function. The percent degradation of dye was determined by decrease in absorbance values of the solution.

Table 4.3: Central composite design matrixes, the predicted and experimental value obtained for the expression of dye degradation percent.

Std	Factor 1 A:Dye uM		Factor 2 B: H_2O_2 M		Factor 3 C:pH		Factor 4 D:time min		Response 1 % degradation [*a]	% dye degradation (square root) [*b]	
										Actual Value	Predicted Value
1	-1	20	-1	0.01	-1	3.75	-1	0	53.17	7.3548	7.7883
2	1	160	-1	0.01	-1	3.75	-1	0	89.74	9.5217	9.0947
3	-1	20	1	0.05	-1	3.75	-1	0	89.82	9.5259	9.3753
4	1	160	1	0.05	-1	3.75	-1	0	75.36	8.7340	8.9760
5	-1	20	-1	0.01	1	10	-1	0	14.93	3.9815	3.8067
6	1	160	-1	0.01	1	10	-1	0	10.85	3.4311	3.4712
7	-1	20	1	0.05	1	10	-1	0	3.85	2.1847	2.6478
8	1	160	1	0.05	1	10	-1	0	0	0.9607	0.6067
9	-1	20	-1	0.01	-1	3.75	1	30	56.79	7.5969	7.9643
10	1	160	-1	0.01	-1	3.75	1	30	91.54	9.6157	9.1392
11	-1	20	1	0.05	-1	3.75	1	30	92.31	9.6557	9.6022
12	1	160	1	0.05	-1	3.75	1	30	77.99	8.8833	9.0715
13	-1	20	-1	0.01	1	10	1	30	19.68	4.5390	4.2836
14	1	160	-1	0.01	1	10	1	30	12.42	3.6528	3.8167
15	-1	20	1	0.05	1	10	1	30	6.56	2.7355	3.1758
16	1	160	1	0.05	1	10	1	30	1.18	1.4502	1.0032
17	-α	20	0	0.03	0	6.875	0	15	22.62	4.8521	3.7820
18	α	160	0	0.03	0	6.875	0	15	4.27	2.2788	3.3489
19	0	90	α	0.01	0	6.875	0	15	16.56	4.1812	4.5099
20	0	90	α	0.05	0	6.875	0	15	16.93	4.2252	3.8966
21	0	90	0	0.03	α	3.75	0	15	88.87	9.4759	9.3522
22	0	90	0	0.03	α	10	0	15	9.34	3.2036	3.3272
23	0	90	0	0.03	0	6.875	α	0	15.3	4.0277	3.9554
24	0	90	0	0.03	0	6.875	α	30	16.46	4.1693	4.2416
25	0	90	0	0.03	0	6.875	0	15	11.29	3.4947	3.9338
26	0	90	0	0.03	0	6.875	0	15	18.2	4.3729	3.9338

[a] Experimental % degradation

[b] Percent degradation that have been transformed according to the requirement of the statistical analysis

The result was analyzed using analysis of variance (ANOVA) as appropriate to the experimental design used. The regression Equation 4.2 was obtained from analysis of variance and all terms regardless of their significance are included in the following equation:

$$\text{Sqrt}(\% \text{ degradation} + 0.92) = 3.66 - 0.22x_1 - 0.31x_2 - 3.01x_3 + 0.14x_4 - 0.43x_1x_2 - 0.41x_1x_3 - 0.03x_1x_4 - 0.69x_2x_3 + 0.013x_2x_4 + 0.075x_3x_4 - 0.37x_1^2 + 0.27x_2^2 + 2.41x_3^2 + 0.16x_4^2 \quad (4.2)$$

A square root transformation was performed with quadratic as original design model which consisted of 1 offset, 4 linear, 4 quadratic and 6 interaction terms. The transformation was required as the Box-Cox plot for power transformation recommended the response to be transformed into square root basis (Figure 4.9) with lambda (λ) value of 0.5 and k-value of 0.9231. This was done to stabilize the variance of the response data as well as greatly improved the appearance of the residual plots.

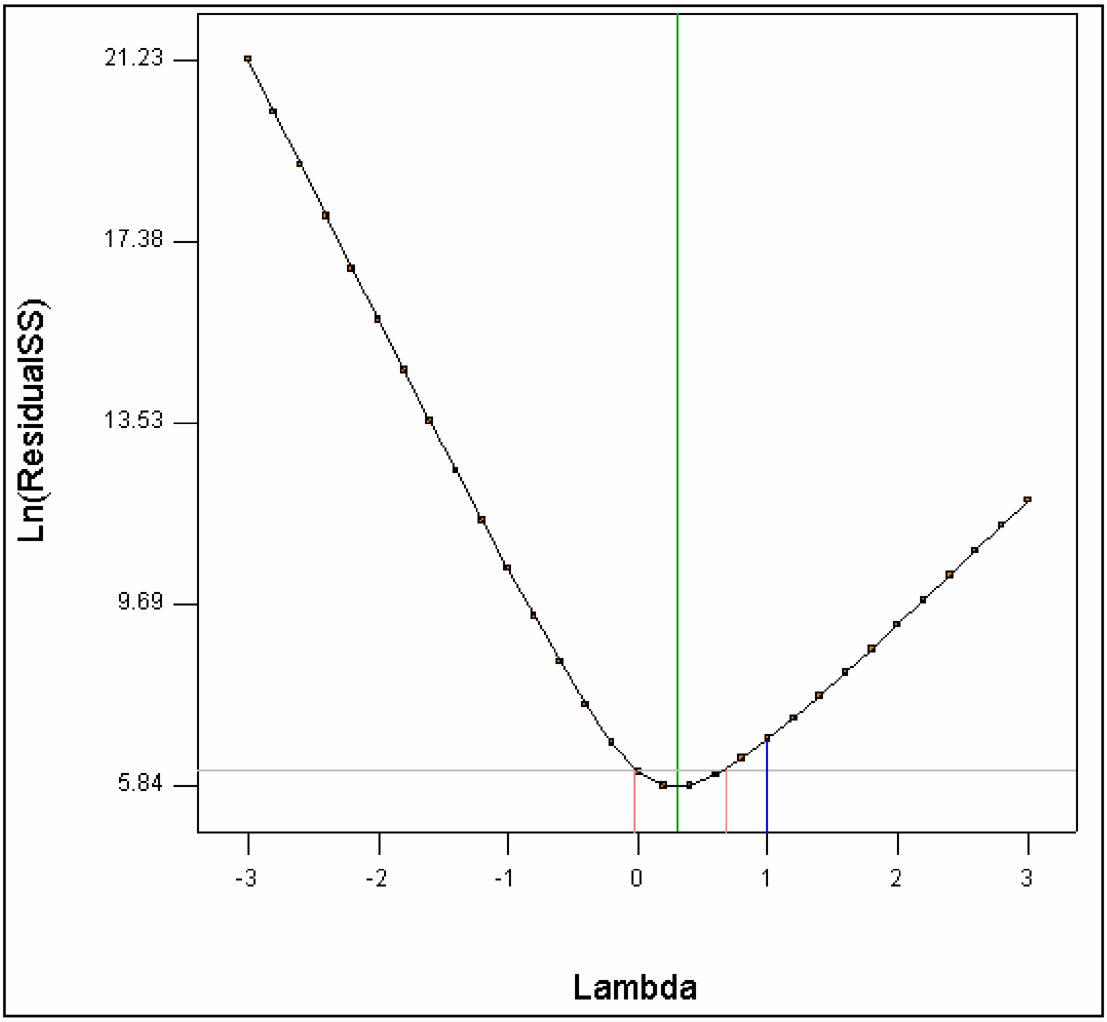

Figure 4.9: The Box-Cox plot of power transformation for AR 94 percent degradation data before data's transformation

The blue line shows the current transformation with λ value of one which symbolized the power applied to the response values. The green line indicates the best λ value which is 0.3, while the red lines indicate the 95% confidence interval. Since the blue line was not in the red line interval, thus the model was not satisfied with desirable response. After data's transformation taken placed, the response model was satisfied with 95 % confidence interval.

Overall result still shows that the 11th run gave the highest percentage of AR 94 degradation which produced 92.31 percent decolorization. The experimental conditions of 11th experiment run are: 20μM of AR 94 concentration, 0.05M of H_2O_2 concentration, pH of 3.75, and 30 minutes of UV irradiation. In addition, there was no photodegradation process at all at run number eight with experimental condition of 160μM, 0.05M, 10, and 0 of AR 94, H_2O_2 concentration, pH value and irradiation time respectively. Thus, there was no optimal point in the selected factors range values.

The coefficient values of Equation 4.2 calculated using Design Expert Software and *P*-value of every term and the interaction are listed in Table 4.3. Based on Table 4.3, the linear term of pH value (x_3), squared terms of pH value (x_3^2), interaction term of AR 94 concentration and H_2O_2 concentration (x_1x_2), interaction term of AR 94 concentration and pH (x_1x_3) and interaction term of H_2O_2 concentration and pH value (x_2x_3) are significant model terms that influence the percent of AR 94 decolorization due to the *P*-value less than 0.05. The other terms seem to be insignificant.

Table 4.4: Regression coefficients and *P*-value calculated from the model

Variables	**Coefficient**	***P*-value[a] (Prob > F)**
Offset	3.660	
x_1 (AR 94 concentration)	-0.220	0.2073
x_2 (H_2O_2 concentration)	-0.310	0.0853
x_3 (pH)	-3.010	< 0.0001
x_4 (time)	0.140	0.3938
x_1^2	-0.370	0.6006
x_2^2	0.270	0.7008
x_3^2	2.410	0.0054
x_4^2	0.160	0.8139
x_1x_2	-0.430	0.0313
x_1x_3	-0.410	0.0367
x_1x_4	-0.030	0.8509
x_2x_3	-0.690	0.0024
x_2x_4	0.013	0.9418
x_3x_4	0.075	0.6681

[a] Values of *P*-value less than 0.0500 indicate model terms are significant

Table 4.4 shows the ANOVA and regression analysis for the photodegradation of Acid Red 94. The precision of a model is indicated by the determination coefficient (R^2) and correlation coefficient (R). The determination coefficient (R^2) implies that the sample variation of 97.60% for photodegradation process was attributed to the independent variables tested. The R^2 value also indicates that only 2.40% of the total variation was not explained by the model. Normally, a regression model having an R^2 value higher than 0.9 is considered to have a very high amount of reduction in the y variability implies the model is good enough. However, a large value of R^2 does not necessarily imply that the regression model is good. This is because adding a variable to the model will always increase R^2, regardless of whether the additional variable is statistically significant or not. But adjusted R^2 statistic will not always increase as variables are added to the model. Thus, it is more suitable to apply adjusted R^2 value to measure the model adequacy.

The adjusted R^2 (coefficient of determination) was calculated to be 94.25%. This value indicates a good agreement existed between the experimental and predicted values of photodegradation of AR 94. When testing the significance of the regression model it was found that *P*-values obtained were small, <0.0001 (Table 4.3) compared to a desired significance level, 0.05. This indicates that the regression model was accurate in predicting the pattern of significance to the AR 94 decolorization.

To investigate the effects of the four factors on percent of AR 94 degradation, the response surface methodology was used and the three-dimensional plot was drawn. The following figures are the response surface curves for the four variables in the photodegradation of AR 94. The response surface representing the percent of degradation was a function of two experimental conditions with the other two conditions being at their optimal levels. It was easy and convenient to comprehend the interaction between two experimental conditions and also to locate their optimum levels.

The AR 94 concentration gave the significant effect to the expression of percent degradation (Figure 4.10). Increase in AR 94 concentration increases the photodegradation process while the lower AR 94 concentration decreases the percent degradation. The maximal percent degradation activity was obtained when the AR 94 concentration was about 90 μM.

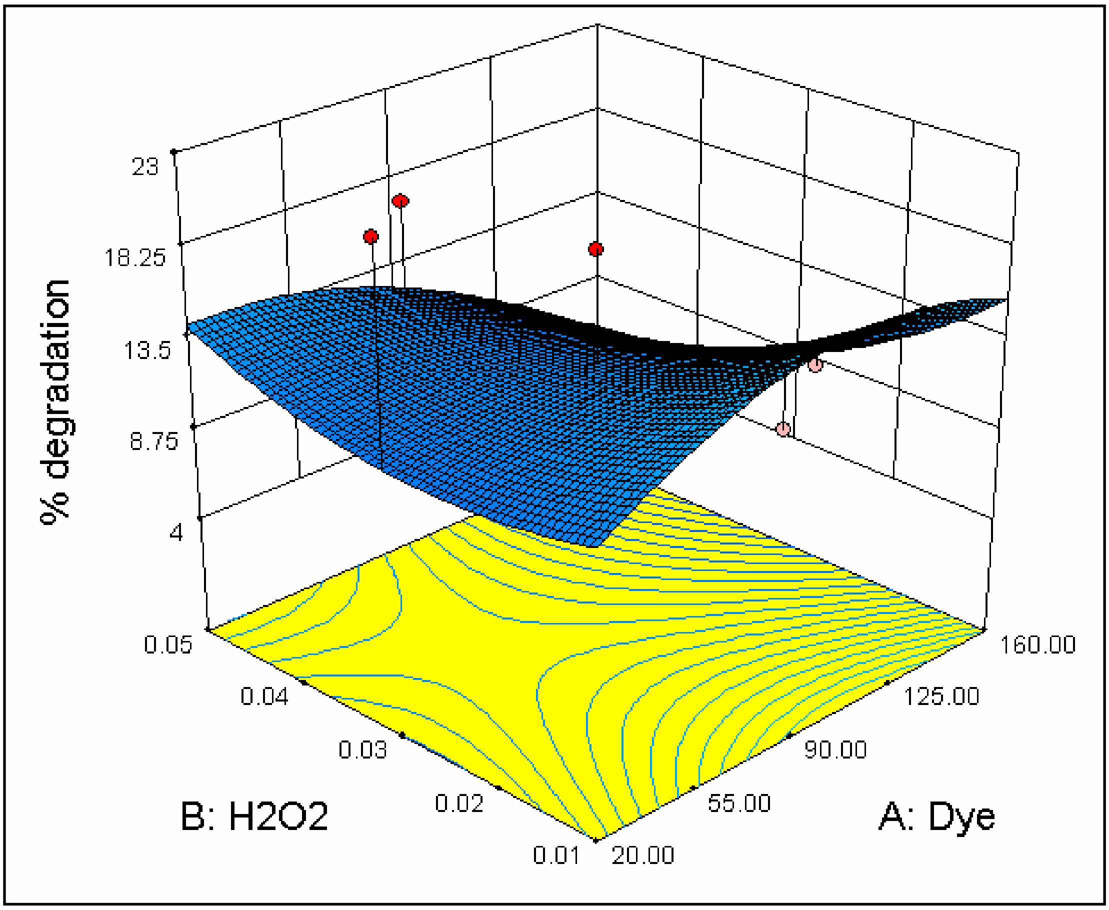

Figure 4.10: Response surface plot of percent degradation: dye concentration vs. H_2O_2 concentration with constant level of pH and time (minutes).

AR 94 concentration made a little impact on the percent dye degradation since maximum percent just lies between 4 to 18 percent. The dye decolorization percent increased up to 90 μM and above this value decreased with increased in AR 94 concentration. However, many dye decolorization studies show that effect of dye concentration was the opposite for response. It is likely the maximum dye decolorization happen to be under low dye concentration (Abdullah *et al.*, 2007), (Körbahti and Rauf, 2008a), and (Körbahti and Rauf, 2008b).

Throughout the experiments, pH seems to give highly significant effect towards percent of dye degradation. When incremental amount of HCl was added into AR 94 solution, decolorization process took place almost immediately without any UV irradiation. This effect is shown in Figure 4.11 in which highly acidic condition gave the highest percent of degradation.

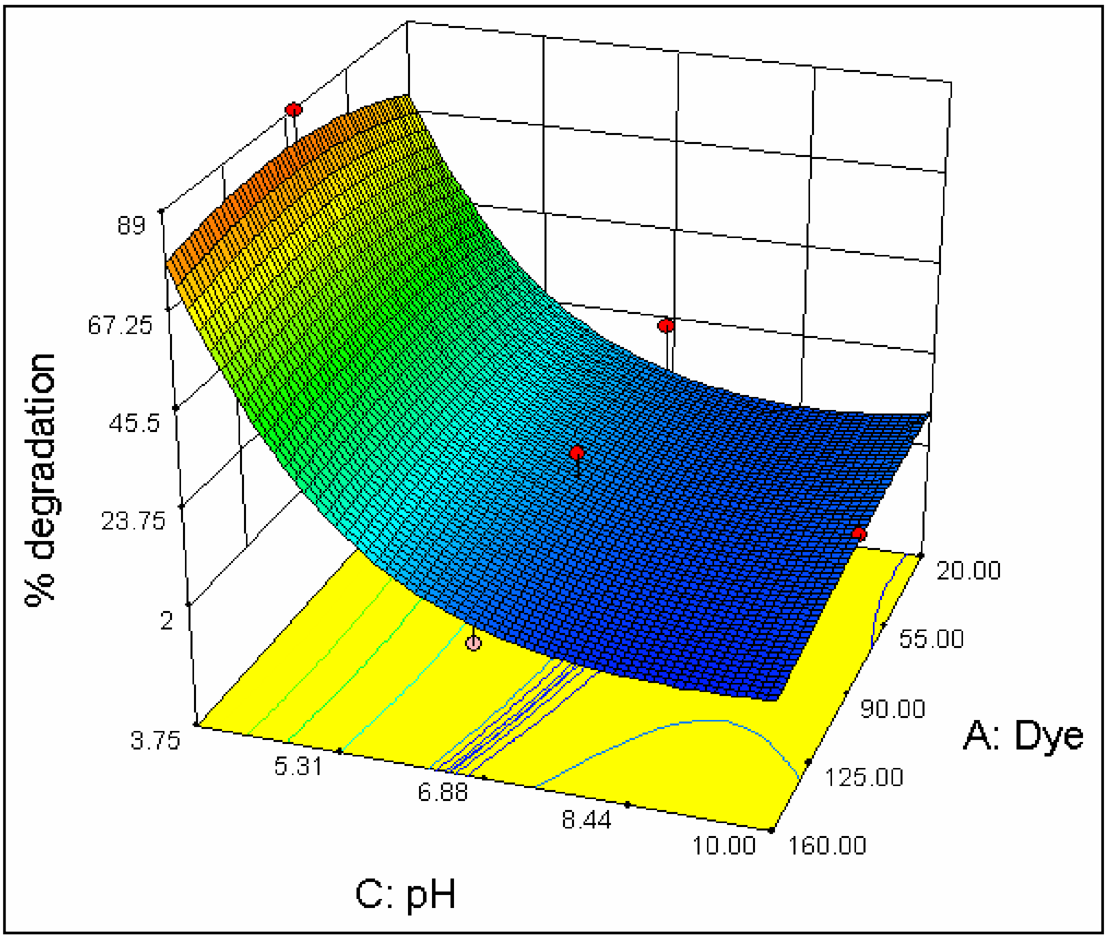

Figure 4.11: Response surface plot of percent degradation: dye concentration vs. pH with constant level of H_2O_2 concentration and time.

The effect of pH value also was studied by Rauf *et al*. (2008), by adding incremental amount of either HCl or NaOH to Acid Red 94 solution in the presence of UV/H_2O_2. They found that the photolytic dye decolorization appears to be less in more acidic solution and increases to a maximum pH of 6.6 and then again starts decreasing in alkaline media. This result was supported by the fact that the enhancement most likely due to peroxide anions (HO_2^-) are produced in solution by UV radiation, which in turn can generate more OH radicals (Aleboyeh *et al*., 2005).

Similar effect has been observed for the interaction effect of pH value against time (Figure 4.12). Although there was no maximum point but there is minimum point of interaction between the factors. The minimum percent of degradation is when irradiation time of eight minutes and pH value of 9.

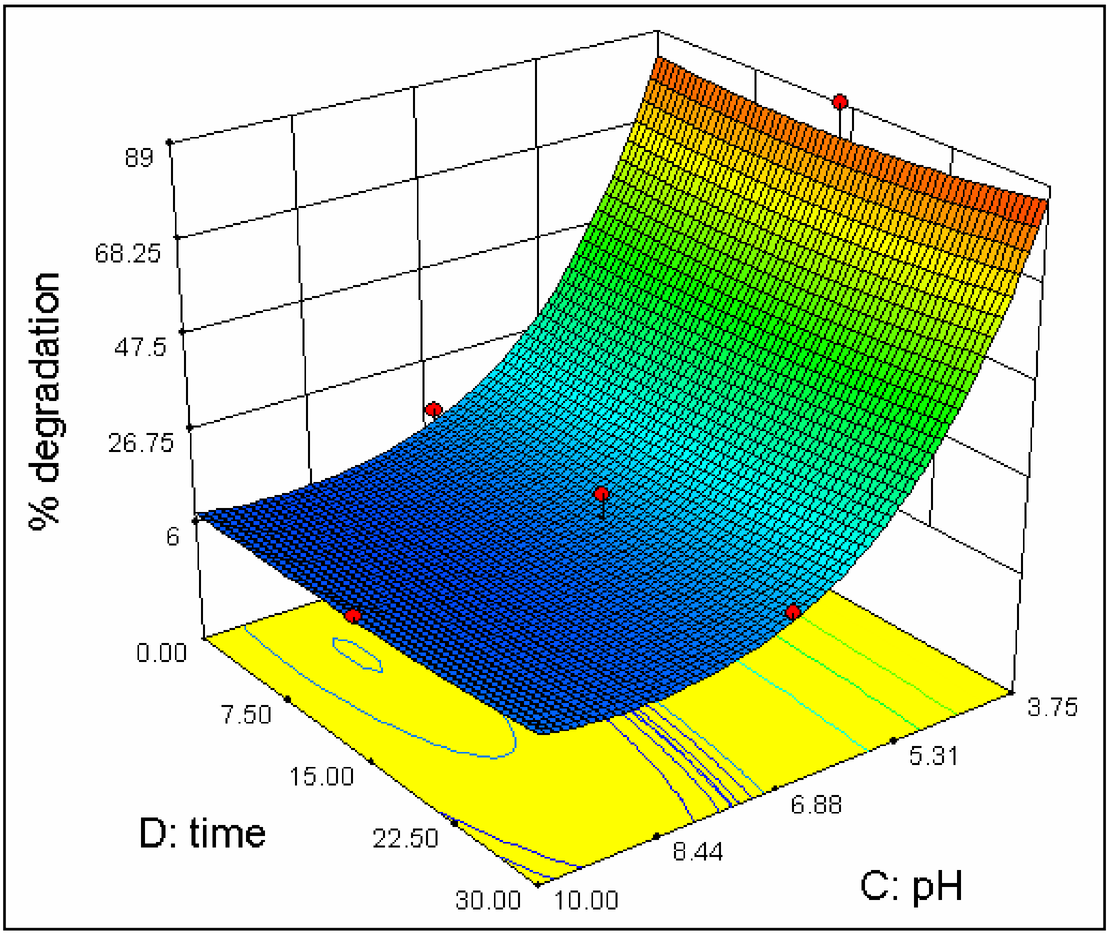

Figure 4.12: Response surface plot of percent degradation: pH vs. time with constant level of AR 94 concentration and H_2O_2 concentration.

Meanwhile, Figure 4.13 illustrates the comparison between all the factors effect at the optimal run conditions in design space. In addition, the plot also shows how the response changes as each factor moves from the chosen reference point, with all other factors held constant at the reference point. The reference point was set by Design Expert software default at the middle of the design space; coded as zero.

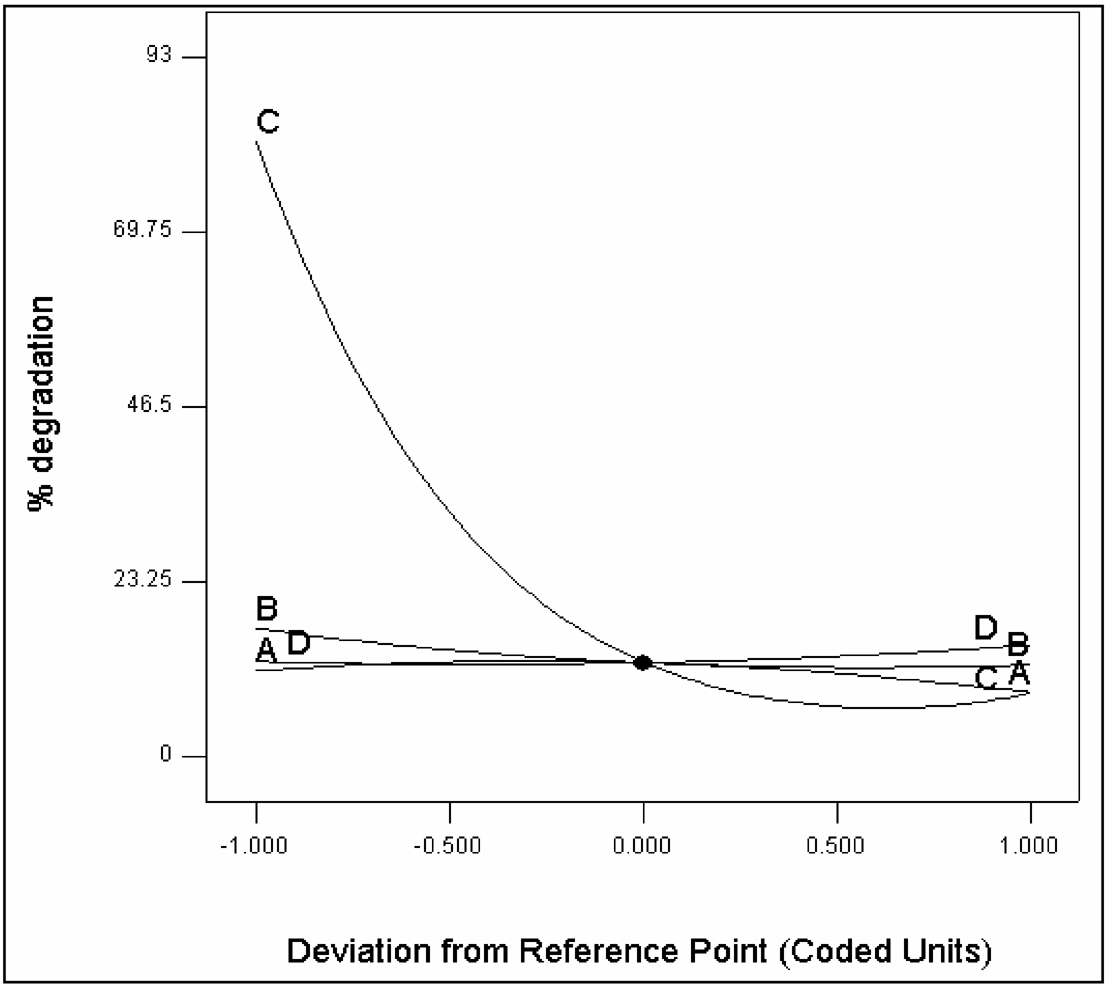

Figure 4.13: Perturbation plot for dye decolorization

However, the optimal point cannot be determined since there was only one significant factor for the desired response. The steep curvature in pH value shows that the response of dye decolorization was very sensitive to this factor. Without any other curvature for the percent of degradation, it can be concluded that there can not be any optimization process to take place.

CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

The best conditions of Acid Red 94 obtained for highest percent of AR 94 decolorization was 20 μM AR 94 concentration, 0.05 H_2O_2 concentration, pH 3.75, and 30 minutes of irradiation time. Meanwhile, the maximum percent of AR 94 decolorization at the best condition was 92.31%.

However, the photodegradation of AR 94 cannot be optimized since there's only one significance factor affecting the desirable response. This is because; AR 94 decolorization was very sensitive to the independent factor of pH.

The proposed model for Central Composite Design fitted very well with the experimental data with R^2 and adjusted R^2 correlation coefficients of 0.9639 and 0.9491, respectively. Thus, the apparent rate of decolorization was found to fit the quadratic model of equation. Effect of independent factors; AR 94 and H_2O_2 concentration, pH, and time have been successfully carried out in this study. Although, the objective of this experiment which to optimize the AR 94 decolorization has not been achieved but the study using response surface methodology (RSM) is well understood.

5.2 Recommendation

Throughout the experiment process, there was much limitation on experimental condition that may end up in significant influence on the desired response. These limitations also stricken the experiment processes to be in the optimum environment. The most obvious limitation in this experiment is inappropriateness of laboratory apparatus used. The UV source for example should be supplied by UV lamp. However, the only UV source that present is just from the laminar flow. Although, there was not any study subjected to this kind of factor but it is believed that along with the other unfavorable environment, there will be a significant effect on the photolytic dye decolorization process.

In addition, the non-stirring condition also limits the dye decolorization process. This is due to the fact that a non-stirred condition will led to uneven UV irradiation. The existence of cloudy solutions after UV irradiation proved that the degradation process was not applied to the whole solutions. This means, only certain part of the solutions were subjected under UV irradiation and only certain areas undergo a complete photo-oxidation process.

Unfortunately, there was also many problems occurred during the experimental period. One of the biggest problems is the unconsistent reads of pH value by pH meter.even though it has been calibrated many times. Thus, it is very hard to control the pH value of the solutions and the probability of an error could occur is high when this process take place.

Thus, it is highly recommended to use a photolytic reactor or well insulated photo-catalytic process. This is because the noise factor can be well managed and enabled the process in its favorable conditions. The irradiation area also will be increased as well.

In addition, the use of real effluent containing dyes will be more suitable as the application of photo-catalytic process take place. At the same time, treated samples can be tested with various tests such as TOC or COD in order to get a better view on the photodegradation of dyes efficiencies. Later on, this research should be further studied to improve the dyes decolorization as high as 100% since there were other powerful oxidative agents present.

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