UNIVERSITI PUTRA MALAYSIA

REMOVAL OF COLOUR FROM PULP AND PAPER MILLS EFFLUENT USING ENZYMATIC TREATMENT AND ADVANCED OXIDATION PROCESSES

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By

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Faculty: Engineering

This research is investigated the efficiency of Advanced Oxidation Processes (AOP) (Fenton and photo-Fenton processes), enzymatic treatment, and combined enzymatic/AOP sequences on colour removal of soda and Chemical Mechanical Pulping (CMP) effluent samples from two major pulp and paper mills in Iran. In addition, the effect of AOP as a pre-treatment unit and post-treatment unit in conjuction with biological treatment, in the combined treatment system, were investigated.

Results indicated that, under all circumstances, Fenton process using UV irradiation (photo-Fenton) was found to be more efficient in the degradation of effluent components, compared to the dark reaction.

Regarding the enzymatic treatment, two kinds of fungal enzymes; Laccase (EC: 1.10.3.2) from *Terametes Versicolour* and Versatile Peroxidase (EC: 1.11.1.7) from *Bjerkandera adusta* were selected and used. In order to determine the effect of enzyme dosage on the overall efficiency of decolourization, experiments were carried out at 2 dosages (1 mg & 2 mg) for each enzyme. To evaluate the effect of external mediator on the enzyme based degradations, each enzyme was applied on the effluent samples, one with the presence of external mediator, and the other, without it, while the other conditions were the same.

It was found that both VP from B*jerkandera adusta* and Laccase from *Terametes versicolour* decolourized the deep brown effluent to a clear light yellow solution. Findings indicated that, an increase in the amount of enzymes (for both VP and Laccase) does not considerably affect the lignin degradation, and resulting in a decrease in the decolourization yield.

In the Laccase treatment process, the decolourization rates of both effluents were enhanced in the presence of ABTS (2, 2′-azino-bis (3-ethylbenzthiazoline-6-sulfonic acid)), while, in case of VP, Mn (II) decreased the efficiency of the treatment.

In combined treatment, biological-chemical sequences (VP/L/Photo-Fenton, VP/L (ABTS) /Photo-Fenton) and, chemical-biological sequences (Photo-Fenton/VP/L, Photo-Fenton/VP/L (ABTS)) were applied.

It has been found that, concomitant use of enzymes and photo-Fenton process produces a considerable effect on colour remediation. The data analysis of sequence treatment indicated that, chemical treatment after the enzymatic stage (photo-Fenton as a post treatment unit) yield a better performance for the CMP effluent; on the contrary, the reverse order (photo-Fenton as a pre-treatment unit) is found to be more efficient for soda effluent.

Keywords:

Advanced Oxidation Processes (AOPs), Colour removal, Enzymatic treatment, Fenton and photo-Fenton processes, Laccase, Pulp and paper mill effluent, Versatile Peroxidase.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

PENGHILANGAN WARNA DARIPADA EFLUEN KILANG PULPA DAN KERTAS MENGGUNAKAN RAWATAN ENZIM DAN PROSES PENGOKSIDAAN TERMAJU

Oleh

SAMANEH KARIMI MAZRAEHSHAHI

January 2009

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Penyelidikan ini mengkaji keberkesanan bagi Proses Pengoksidaan Termaju (AOP) (proses Fenton dan photo-Fenton), rawatan enzim, dan gabungan turutan enzim/AOP ke atas penghapusan warna bagi sampel efluen soda dan pempulpaan kimia mekanikal (CMP) daripada dua kilang pulpa dan kertas utama di Iran. Selanjutnya, kesan AOP sebagai unit pra-rawatan dan unit pasca rawatan di dalam rawatan gabungan juga dikaji.

Keputusan menunjukkan bahawa, dalam semua keadaan, proses Fenton yang menggunakan pancaran UV (photo-Fenton) didapati lebih berkesan dalam mendegradasi komponen efluen, dibandingkan dengan tindakbalas gelap.

Mengenai rawatan enzim, dua jenis enzim, iaitu Laccase (EC: 1.10.3.2) daripada *Terametes Versicolour* dan Versatile Peroxidase (EC: 1.11.1.7) daripada *Bjerkandera adusta* telah dipilih dan digunakan. Untuk mengetahui kesan pengedosan ke atas keberkesanan mengenyahwarna secara keseluruhan, kajian telah dilakukan pada dua dos (1 mg dan 2 mg) bagi setiap enzim. Untuk menilai kesan mediator luaran ke atas pendegradasi yang berasaskan enzim, setiap enzim digunakan ke atas sampel, satu dengan kewujudan mediator, dan satu lagi tanpa mediator, sementara keadaan lain adalah sama.

Didapati bahawa kedua-dua VP daripada B*jerkandera adusta* dan Laccase daripada *Terametes versicolour* menukarkan warna efluen daripada cokelat pekat kepada satu larutan kuning terang. Hasil kajian menunjukkan bahawa, pertambahan kuantiti enzim (bagi kedua-dua VP dan Laccase) tidak begitu menjejaskan pendegradasi lignin, dan pengurangan dalam mengenyah warna. Di dalam proses rawatan Laccase, kadar pengurangan warna bagi kedua-dua efluen ditingkatkan dengan kewujudan ABTS (2, 2'-azino-bis (3-ethylbenzthiazoline-6-sulfonic acid)), sementara, dalam kes VP, Mn (Π) telah mengurangkan keberkesanan rawatan.

Dalam rawatan gabungan, turutan biologi-kimia (VP/L/Photo-Fenton, VP/L (ABTS) /Photo-Fenton) dan, turutan kimia-biologi (Photo-Fenton/VP/L, Photo-Fenton/VP/L (ABTS)) telah digunakan. Didapati bahawa, penggunaan kedua-dua proses enzim dan photo-Fenton secara

beriringan menghasilkan kesan ketara ke atas penghilangan warna. Analisis data rawatan berturutan menunjukkan bahawa, rawatan kimia selepas tahap enzim (sebagai unit pasca rawatan photo-Fenton) menghasilkan prestasi yang lebih baik bagi efluen CMP; sebaliknya, turutan berlawanan (photo-Fenton sebagai unit pra-rawatan) didapati lebih berkesan bagi efluen soda.

Katakunci:

Efluen pulpa dan kertas, Laccase, Penghilangan warna, Proses Fenton dan photo-Fenton, Proses Pengoksidaan Termaju (AOP), Rawatan Enzim, Versatile Peroxidase,

DEDICATION

This dissertation is dedicated

to my dear...dear uncle which I cannot find any word in this world to express my

sincerest appreciation to him

to my parents for their love and patience

to Arash & Lida, Afshin & Maryam and to lovely Ramona

….your essence is in everything I accomplish

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LIST OF ABBREVIATIONS

Abbreviations	Meaning
ABTS	2,2´-azinobis(3-ethylbenzthiazoline-6-sulphonate)
Abs	Absorbance
AOX	Adsorbable Organic Halogen
AOP	Advanced Oxidation Process
BOD	Biological Oxygen Demand
B. adusta	*Bjerkandera adusta*
CMP	Chemical Mechanical Pulping
COD	Chemical Oxygen Demand
CTMP	Chemi-Thermo Mechanical Pulping
CU	Colour Unit
ECF	Elemental Chlorine Free
EPA	Environmental Protection Agency
F	Fenton
GPC	Gel Permeation Chromatography
HPLC	Hhigh Performance Liquid Chromatography
kDa	kiloDalton
L	Laccase
LME	Lignin Modifying Enzymes
LiP	Lignin Peroxidase

MnP	Manganese Peroxidase
MVR	Mechanical Vapor Recompression
NB	None Biodegradable
NHE	Normal hydrogen Electrode
P. chrysosporium	Phanerochate Crysosporium
Ph	photo-Fenton
Pt-Co	Platinum Cobalt
PAC	Poly Aluminum Chloride
PAH	Poly Aromatic Hydrocarbons
RB	Ready Biodegradable
SS	Suspended Solid
T. versicolour	Terametes versicolour
TMP	Thermo Mechanical Pulping
TSS	Total Suspended Solid
TCF	Totally Chlorine Free
UV	ultra Violet
VA	Veratryl Alcohol
VP	Versatile Peroxidase
VOC	Volatile Organic Compound
WRF	White Rot Fungi

CHAPTER 1

INTRODUCTION

1.1 Background

The pulp and paper industry is the third largest water consuming industry in the world. This industry generates a crucial perplexity in terms of environmental pollution as a consequence of its black liquor effluent. Until recently, colour was not considered to be a major problem, being classified as a non-conventional pollutant (Ali and Sreekrishnan, 2001). Even so, it has now, been known that the discharge of coloured effluent from pulp and paper mills is not only a serious aesthetic problem, but also has other critical problems.

This industry generates a high coloured effluent, as a result of several stages in processing of wood and pulp which include wood digesting, washing, bleaching and drying of pulp. This colour is mainly attributed to the complex compounds derived from lignin degradation products (Kreetachat et al. 2006).

1.2 Importance of Research

At the present time one of the pivotal concerns of science is water pollution control and wastewater treatment. In this aspect treatment of coloured-

effluents remains a problem. As Lucas and Peres (2005) reported, coloured wastewaters are a large problem for conventional treatment plants in all part of the world. Based on this, decolourization of industrial effluents is one of the major scientific interests in recent years.

Colour is the most visible contaminant in the wastewater. Just a little amount of colour in water (10-50 mg/L) is highly unpleasant and deteriorates aesthetic aspects, glassiness and gas solubility of water bodies (Wong and Yu 1999). In addition, colourant compounds are responsible for problems such as mutagenic and carcinogenic activities (Chung and Stevens 1992). The other critical problem associated with colour is reduction in the photic depth of the receiving water, on the other hand, decreasing in the penetration of solar radiation. It causes a marked change in the natural photosynthesis process of water bodies, adverse effect on aquatic eco-system and its productivity (Ramanathan, 1989; Sahoo and Gupta 2004).

Colour removal from wastewater is often more consequential than the removal of soluble colourless organic substances which usually contribute the major fraction of chemical or biologicall oxygen demand (Banat et al. 1996).

1.3 Problem Statement

The volumetric load of industrial effluent discharge to neighboring water bodies has an ascending rate. It brings serious health concerns to environmental regulatory agencies. Therefore, several countries have imposed specific colour levels for discharging effluent by means of strict environmental legislation. Nevertheless, up to now no economically sound process has been found.

Aerated lagoons and activated sludge plants are the most common systems used in pulp and paper wastewater treatment. By means of such conventional systems appreciable reduction in biological oxygen demand (BOD), total suspended solids (TSS), and critical toxicity could be achieved, but in terms of colour reduction are not effective. As indicated in all literatures these systems are not functional for treating colourful effluents.

Rana et al., (2004) stated that; the most widely used biological treatment system is activated sludge process; however, it also is ineffective in colour removal of the effuents. Joyce and Petke (1983) reported that; commonly aerated lagoons or activated sludge units can eliminate conventional pollutants such as BOD and COD at efficiencies of up to 95%, while these biological systems are capable of colour reductions of only 0-30% (Rush and Shannon, 1976, and Obiaga and Ganczarczyk, 1974). This resistance of colour bodies to biological treatment is largely due to the inability of the microbial