UNIVERSITI TEKNOLOGI MARA

EFFECT OF TEMPERATURE TOWARDS TYROSINASE ENZYME IMMOBILISED ON MAGNETIC CHITOSAN

MUHAMMAD AZMEER ASHRAF BIN KAMAL ARIPIN

Dissertation submitted in partial fulfilment of the requirements needed for the degree of **Bachelor of Engineering (Hons) Chemical and Bioprocess**

FACULTY OF CHEMICAL ENGINEERING UNIVERSITI TEKNOLOGI MARA SHAH ALAM

July 2017

ACKNOWLEDGEMENT

First and foremost, I would like to express my gratitude to Allah S.W.T, The Almighty God for giving me His guidance during the process of completing my studies. Alhamdulillah, He gave me the strength, knowledge, ability and opportunity to undertake this research.

I also want to thank both of my parents for giving me their undying love and moral support during the research program. All of my progresses in my academic years have all been solely for them.

My deep gratitude goes to Dr. Nik Raikhan Binti Nik Him for sharing with me her vast knowledge on enzymes and their application. Her unwavering enthusiasm and passion on biotechnology application kept me constantly engaged with my research and also her personal generosity helped me in numerous times at UiTM. I would also like to give my sincere appreciation to my co-supervisor, Mohammad Fiqri bin Zainuddin, for enlightening me with his knowledge on biosensor application and his help during my experiment.

My indebtedness also extends to laboratory assistants for helping me with acquiring the raw material and using the equipment. Last but not least, thank you to my peers who have helped in generating valuable ideas during group discussions whenever I meet dead-ends. Without their contributions, I might not be able to finish with my study.

ABSTRACT

Bisphenol A (BPA), a compound that is traceable in wastewater and packaging material can bring detrimental effect to human health when it is consumed. BPA has been linked to particular diseases related to endocrine system, such as breast cancer among women and defective reproductive system among men. In this modernised world, there is limitation of method to detect the presence of the compound in the environment as the available techniques are usually expensive, cannot be conducted insitu, and require experienced technicians to operate effectively. In this study, the tyrosinase enzyme was employed to identify the presence of the BPA compound and to degrade the BPA into its monomer because the method of preparation is simple and relatively cheap compared to other existing techniques used to identify the presence of BPA in a solution. In this research, only effect of temperature was studied towards the ability of the immobilised tyrosinase enzyme to degrade the phenolic compound. The tyrosinase enzyme was immobilised onto magnetic chitosan via simple crosslinking process with the aid of glutaraldehyde as the crosslinking medium. Other parameters that influence the enzyme behaviour, like pH, hydrophobicity and electrostatic interactions, were not investigated due to time constraints and lack of necessary equipment and assumed to be similar from one set of experiment to another by maintaining stirring rate, amount of chemicals used in preparation steps. The percentage of biodegradation of BPA using immobilised tyrosinase enzyme on magnetic chitosan compound under different temperatures ($T = 20,25,30,35,40^{\circ}C$) was determined using spectrophotometry analysis at 465nm of wavelength. Based on the results obtained, the tyrosinase managed to degrade highest amount of BPA at temperature, $T = 20^{\circ}C$.

Keywords — Tyrosinase, magnetite chitosan, enzymes immobilisation, bisphenol A, temperature

TABLE OF CONTENTS

	PAGE
AUTHOR'S DECLARATION	ii
CERTIFICATION	iv
ACKNOWLEDGEMENT	vi
ABSTRACT	vii
TABLE OF CONTENTS	viii
LIST OF TABLES	xi
LIST OF FIGURES	xii
LIST OF SYMBOLS	xiv
LIST OF ABBREVIATIONS	XV

CHAPTER 1 INTRODUCTION

1.1	Research Background	1
1.2	Problem Statement	2
1.3	Objectives of Research	3
1.4	Scopes of Research	3

CHAPTER 2 LITERATURE REVIEW

2.1	Introduction to Catalyst		4
	2.1.1	Basic Principles of Enzymes	5
2.2	Tyrosinase Enzyme as a Component of Biosenson		10
	2.2.1	Tyrosinase Enzyme Background	10
	2.2.2	Bisphenol (BPA) Compound Background	12

CHAPTER ONE

INTRODUCTION

1.1 RESEARCH BACKGROUND

People generally know that the Earth is estimated to contain about of 75% water. Out of this percentage, the amount of water available for consumption is very limited due to several factors, mostly due to inaccessibility of water (Peavy *et. al.*, 1985). What if this proportional amount of water is contaminated with hazardous agents that can risk your health and your comfortability in living very significantly? What if these life threatening compounds are already inside of the living organisms' bodies due to using certain objects to carry out their regular routine?

There are so many adverse effects that can occur if the presence of these harmful chemicals in the environment is left untreated to an extent of time. Many concerns have been raised to demand a limitless supply of clean resources and consequently, many approaches have been vigorously proposed and conducted to minimise the effects of these toxicants. However, the current technology that can detect these harmful compounds before any tests can be performed to analyse the most suitable method to minimise the dangerous chemicals has many limitations. One of the novel applications to bring the gap between lack of technology to detect water pollutants and the act of augmenting the water quality closer is the invention of biosensor.

A biosensor is defined as the device that incorporates a bio-receptor and a transducer components to detect specific compounds and monitor the presence, reaction activity and concentration of the analytes. The bio-receptor is a biomolecule that binds the desired and specific chemical substance while the transducer will aid in converting the recognition event into a measurable signal. In this research, the enzyme that acts as bio-receptor is the immobilised tyrosinase enzyme, which its ability to degrade phenolic compounds will be investigated under effect of temperature. Whereas, the target substrate that will be used for the catalytic reaction is bisphenol A (BPA) compound.