

UNIVERSITI TEKNOLOGI MARA

WILD-TYPE *Escherichia coli*
DISINFECTION BY FLOW CELL
ULTRASOUND: MECHANISMS AND
OPTIMISATION THROUGH
NEURAL NETWORKS

NORAINI BINTI MAT BUDARI

Thesis submitted in fulfilment
of the requirements for the degree of
Doctor of philosophy
(Civil Engineering)

College of Engineering

March 2022

ABSTRACT

The goal of this thesis is to investigate the use of the flow cell ultrasonic reactor as a disinfection treatment for the disruption of wild-type cells of *E. coli* as model organisms and to recommend that the custom ultrasonic reactor be adaptable to real environmental disinfectants as there is growing global concern about green disinfection technology. There are several challenges associated with ultrasound treatment; the requirement for high energy consumption continues to be a significant constraint on ultrasonic application in water and wastewater treatment systems, as well as industrial applications. Additionally, the lower cavitation region and irradiating surface of the ultrasonic reactor reduce treatment performance. Besides that, there is insufficient information about the cell's mechanical impacts, as evidenced by qualitative and quantitative methods using depth microscopy and particle size analysis of ultrasonic treatment on *E. coli* wild-type cells disruption. A comprehensive investigation using a combination of microscopy techniques, including gram-staining analysis, Environmental Scanning Electron Microscopy and High-Resolution Transmission Electron Microscopy, demonstrated and concluded that flow cell ultrasound treatment had a significant effect on cell disruption. Furthermore, with the design of frequency transducers at 30 kHz, this improves the performance of the ultrasonic reactor and has a greater impact on the disinfection process. Moreover, the interconnections between operational parameters in flow cell ultrasound treatment have an impact on treatment performance. The optimal operation of an unsubmerged reactor is associated with the geometry of the reactor structural design. The curve estimation confirms that the behaviour of cells inactivated to the treatment duration in multilayer perceptron yielded a regression line with a determination coefficient greater than 89%, together with a lower intercept as model bias fit a data set to the linear regression model behaviour. RMSE has given the average error between the experimental data and the predicted value. In addition, the residuals-predicted on the experimental model were not clear patterns and were considered a good fit model. Hence, this presents greater confidence in the linear regression model behaviour between predicted and observed data. The 60-minute treatment duration and cell inactivation in linear regression behaviour ($p < 0.0001$) and further extended treatment duration until 110 minutes led to the change in the behaviour of quadratic models ($R^2 = 0.811$). The increase in treatment duration with of more than 60 minutes appeared to reduce the death cell rate. The trend removal reduced the rapid disruption and generated a tailing phenomenon. It is important to note that the higher performance of the flow cell ultrasonic reactor on inactivation effect was referred to the sonication parameters. This resulted in the interaction between microbubbles and membrane cells with mechanical and chemical damage inactivation effects. Thus, the mechanical inactivation effects such as liquid microjets, shockwaves, acoustic streaming, and combinations of chemical cytotoxic effects through reactive oxygen species act as a pillar in the cell's inactivation effect. Meanwhile, oxidative radical-induced lipid oxidation was the primary target for membrane oxidation, and it was noted that oxidation was advantageous in a membrane because oxygen was more concentrated in the hydrophobic environment of lipid bilayers than in solution. Further, microscopy images and particle size analysis showed flow cell ultrasound treatment caused lethal effects due to mechanical damage to the cells and a proven tailing phenomenon during the treatment. Therefore, the flow cell ultrasonic reactor was an efficient sustainable disinfection technology towards increasing energy efficiency.

ACKNOWLEDGEMENT

In the name of Allah, the Most Gracious and the Most Merciful. All praises to Allah and His blessing for the completion of this thesis. For all the possibilities and strength while in this journey there've been too many tears falling, I thank Allah with all His sustenance. I experienced so much during this process, not just from the academic perspective, but from the personality side as well. My humblest gratitude to the holy Prophet Muhammad (Peace and Blessings be upon Him) whose way of life has been a continuous guidance for me.

First and foremost, I would like to thank my supervisor sincerely, Prof. Dato' Ir. Ts. Dr. Haji Mohd Fozi Ali for his guidance, understanding, patience and most importantly, he has provided constructive motivation and a warm spirit to finish this thesis. Getting him as my supervisor was a great pleasure and honour. I am very grateful to my co-supervisors as well, Prof. Dr. Ku Halim Ku Hamid for their diligent advice and encouragements. I wish to express my sincere appreciation to Assoc. Prof. Dr. Khalilah Abd Khalil for the help in bacterial enumeration and in using the facilities in the Microbiology Laboratory. This study would not have been the same as discussed here without their continuing assistance and commitment.

I am strongly indebted to Universiti Teknologi MARA (UiTM) together with Ministry of Education Malaysia for providing scholarship Young Lecturer Scheme, Ministry of Higher Education (MOHE) for the grant Research Development Grant Scheme (600-RMI/RAGS 5/3 (151/2014)). Also, Universiti Teknologi MARA (UiTM) for giving me this golden opportunity to pursue PhD; mainly Institute of Graduate Studies (IPSIS), Faculty of Civil Engineering (FKA), Faculty of Chemical Engineering (FKK), Faculty of Applied Sciences (FSG), Institute of Research Management and Innovation (IRMI) and Tun Abdul Razak Library (PTAR).

My deepest gratitude goes to Ts. Dr. Safari Zainal from FKK for the help in research grant scheme and Dr. Rasimah Aripin from TMSK for the guidance in SPSS information. I also offer my special thanks to Mr. Mohibah Musa (FKK), Mr. Hazri Othman (Environmental Laboratory, FKA), Mrs. Nor Ramliza Ramli (BIOREC Research Laboratory, FKA), Mrs. Azizan Din from FKK for her assistance with the particle size analyser testing instrument and also from my MSc and PhD colleague from FKA and FSG among them are Nurul Fatimah Khairuddin, Siti Aisyah Ghazali and all the members in the postgraduate room at level 4, FKA who consciously and unknowingly guided me to an understanding of some of this journey's hidden obstacles and inspiring me to survive until the end.

My heartfelt thanks go to all of my family members. It would be impossible to complete this thesis without their assistance. I'd like to express my gratitude to my parents, the late Mat Budari Darman and Siti Zainap Hanapi, as well as my siblings Sulaila, Hisham and Md Othman, and all my family members, including my parent-in-law Ramlan Yusof and Robiah Selamat and my sisters-in-law. Finally, I'd like to thank my supportive husband, Mohd Fauzi Ramlan, as well as my children Iman Firdaus, Iman Nurdhia Fatimah and Iman Deen Al-Fateh. I'd like to express my deepest gratitude, without your support, motivation and understanding, I might not be able to complete this PhD journey.

TABLE OF CONTENTS

	Page
CONFIRMATION BY PANEL OF EXAMINERS	ii
AUTHOR'S DECLARATION	ii
ABSTRACT	ix
ACKNOWLEDGEMENT	x
TABLE OF CONTENTS	xi
LIST OF TABLES	xvi
LIST OF FIGURES	xviii
LIST OF SYMBOLS	xxviii
LIST OF ABBREVIATIONS	xxix
CHAPTER ONE: INTRODUCTION	1
1.1 Background of the Study	1
1.2 Problem Statement	6
1.3 Objectives of the Study	8
1.4 Scope and Limitation of the Study	8
1.5 Importance of the Study	12
CHAPTER TWO: LITERATURE REVIEW	13
2.1 Introduction	13
2.2 Disinfection	13
2.3 Ultrasound	16
2.3.1 Principle of Ultrasound	17
2.3.2 Mechanism	19
2.3.3 Action of Inactivation Mechanism	20
2.3.3.1 Cavitation Bubbles and Cell Interactions	21
2.3.3.2 Energy Changes during Ultrasound Treatment	27
2.3.4 Reactor Designs of Ultrasound	28
2.3.5 Influencing Parameters of Ultrasound on Microorganism Cell Disruption	33

CHAPTER ONE

INTRODUCTION

1.1 Background of the Study

Water is a necessity in our daily life. It is now universally agreed that providing the community with safe water acts as a preventive measure to prevent outbreaks of waterborne plague. In addition, the demand for clean water supply has risen in the new decade of the twenty-twenties. Due to increased contamination as a result of industrialisation and urbanisation activities, conventional drinking water treatment systems encountered difficulties (Figure 1.1).

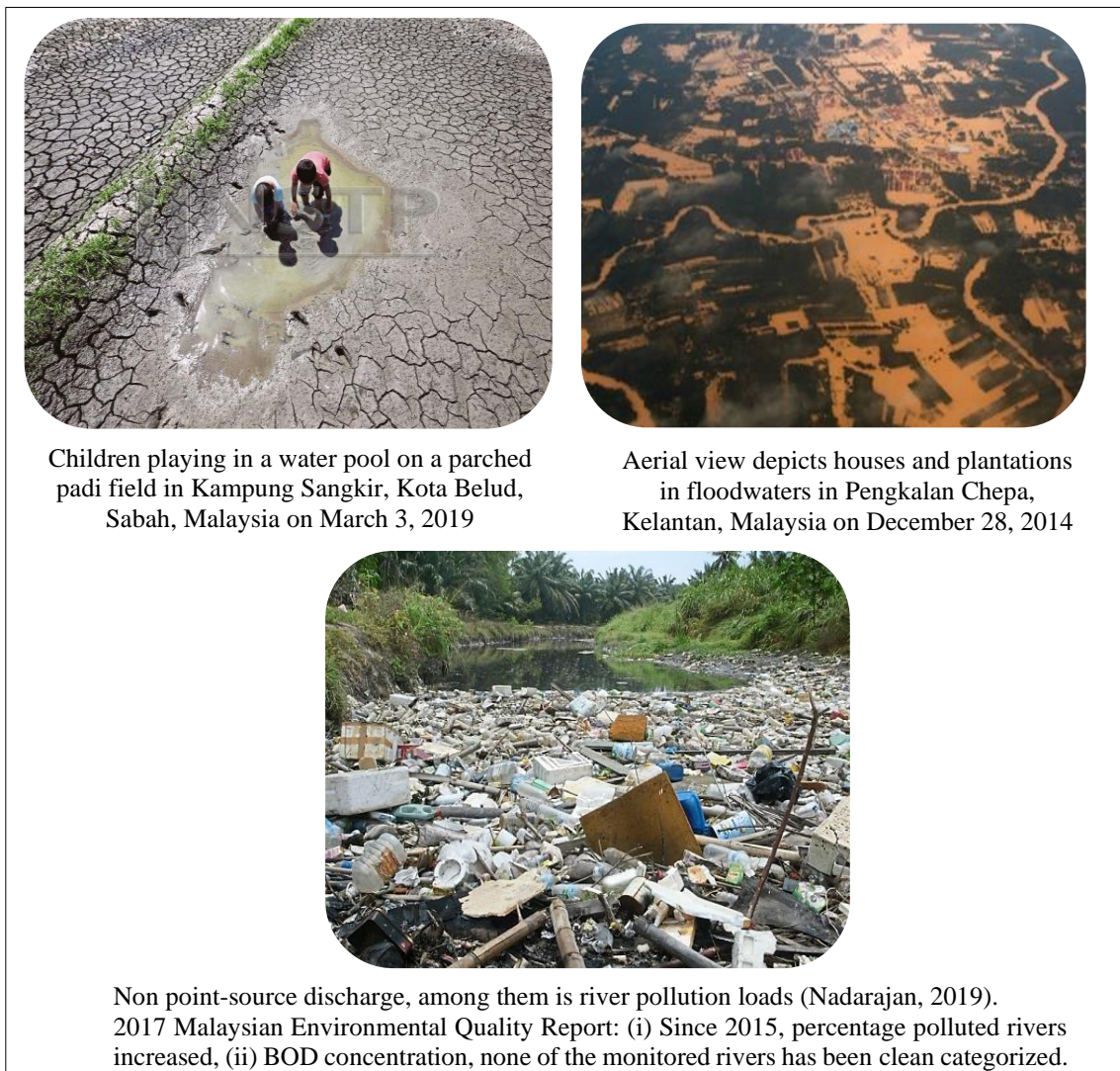


Figure 1.1 Contaminating Sources of Surface Water