BIOACCUMULATION OF RARE EARTH ELEMENT LANTHANUM IN *Pistia stratiotes* (WATER LETTUCE) AND *Oreochromis niloticus* (NILE TILAPIA) IN ARTIFICIAL WASTEWATER SYSTEM

BY

MUHAMMAD NAZREEN BIN MUHAMMAD NOR

A thesis submitted in fulfilment of the requirement for the degree of Master of Health Sciences

Kulliyyah of Allied Health Sciences
International Islamic University Malaysia

FEBRUARY 2019

ABSTRACT

Rare earth elements (REEs) are a series of chemical elements found in the earth's crust that are vital to many modern technologies, including consumer electronics, computers, communications and clean energy technologies. However, during the production of REEs, the activity is susceptible in causing environmental contamination by discharging the REE industrial wastewater into the environment especially ponds, lakes and rivers. The REE wastewater containing Lanthanum (La) residue and other radioactive elements will be uptaken by the aquatic plants and freshwater fishs through bioaccumulation process. Therefore, this study aims to determine the effectiveness of the aquatic plant, i.e. *Pistia stratiotes* and freshwater fish (*Oreochromis niloticus*) as bioaccumulation agents of rare earth metals, especially Lanthanum (La) in REE artificial wastewater system. The study was carried out by subjecting *P. stratiotes* (water lettuce) and *O. niloticus* (Nile tilapia) to 4 levels of Lanthanum Chloride ($LaCl_3$) treatments (*i.e.* 1, 3, 5 and 10 ppm) for 12 days in aquaponics artificial wastewater system. The samples were collected every two days and dried prior to digestion. The dried plant parts (root and leaf) and fish tissues (stomach and flesh) were acid-digested and then analyzed the REE metal (La) accumulation using the Inductively Coupled Plasma-Mass Spectrometry (ICP-MS). Overall results showed REE (i.e. La) was accumulated in the highest amount in the stomach compared to other parts of the plant and fish tissues.

خلاصة البحث

العناصر الأرضية النادرة هي عبارة عن سلسلة من العناصر الكيميائية الموجودة في القشرة الأرضية، والتي تعتبر مهمة للعديد من التقنيات الحديثة، مثل الإلكترونيات الاستهلاكية، وأجهزة الكمبيوتر والاتصالات، وتقنيات الطاقة النظيفة. ومع ذلك فإن استخراج العناصر الأرضية النادرة يكون عرضة للتسبب في التلوث البيئي عن طريق تصريف مخلفاتها السائلة في البيئة، خاصة البرك والبحيرات والأنهار. تأخذ النباتات المائية وأسماك المياه العذبة المخلفات السائلة للعناصر الأرضية النادرة من خلال عملية التراكم الأحيائي والتي تحتوي على رواسب عنصر اللانثانوم وغيرها من العناصر المشعة. ولذلك هدفت هذه الدراسة إلى تحديد مدى فعالية النبات المائي *Pistia stratiotes* (خس الماء) وسمك الماء العذب *Oreochromis niloticus* (البلطي النيلي) كعوامل للتراكم الأحيائي للعناصر الأرضية النادرة، وخاصة عنصر اللانثانوم في نظام صناعي للمخلفات المائية. أجريت الدراسة عن طريق تعريض ماء الخس والبلطي النيلي إلى 4 مستويات من كلوريد اللانثانوم ($LaCl_3$) (1، 3، 5، و 10 جزء في المليون) لمدة 12 يوما في النظام الغذائي المائي للنظام الصناعي للمخلفات المائية. تم جمع العينات كل يومين وتجفيفها قبل عملية هضمها للتحليل. تم تجزئة الأجزاء النباتية المجففة (الجذور والأوراق) وأنسجة الأسماك (المعدة واللحم) بواسطة الأحماض، ومن ثم تحليل عنصر اللانثانوم باستخدام المطياف الكتلي البلازمي بالتقارن الحثي (ICP-MS). أظهرت النتائج الإجمالية أن اللانثانوم قد تراكم في المعدة بأعلى كمية مقارنة بالأجزاء الأخرى من الأنسجة النباتية والسمكية.

APPROVAL PAGE

I certify that I have supervised and read this study and that in my opinion, it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a thesis for the degree of Master of Health Sciences

…………………………………..
Ibrahim Bin Abu Bakar
Supervisor

…………………………………..
Muhammad Nor Bin Omar
Co-Supervisor

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a thesis for the degree of Master of Health Sciences

…………………………………
Shahbudin Bin Saad
Internal Examiner

…………………………………..
Saiful Nizam Bin Tajuddin
External Examiner

This thesis was submitted to the Department of Biomedical Sciences and is accepted as a fulfilment of the requirement for the degree of Master of Health Sciences

…………………………………..
Hanani Binti Ahmad Yusof
Head,
Department of Biomedical Sciences

This thesis was submitted to the Kulliyyah of Allied Health Sciences and is accepted as a fulfilment of the requirement for the degree of Master of Health Sciences

…………………………………..
Suzanah Abdul Rahman
Dean,
Kulliyyah of Allied Health Sciences

DECLARATION

I hereby declare that this dissertation is the result of my own investigations, except where otherwise stated. I also declare that it has not been previously or concurrently submitted as a whole for any other degrees at IIUM or other institutions.

Muhammad Nazreen Bin Muhammad Nor

Signature .... Date ....

INTERNATIONAL ISLAMIC UNIVERSITY MALAYSIA

DECLARATION OF COPYRIGHT AND AFFIRMATION OF FAIR USE OF UNPUBLISHED RESEARCH

BIOACCUMULATION OF RARE EARTH ELEMENT LANTHANUM IN *Pistia stratiotes* (WATER LETTUCE) AND *Oreochromis niloticus* (NILE TILAPIA) IN ARTIFICIAL WASTEWATER SYSTEM

I declare that the copyright holders of this dissertation are jointly owned by the student and IIUM.

No part of this unpublished research may be reproduced, stored in a retrieval system, or transmitted, in any form or by any means, electronic, mechanical, photocopying, recording or otherwise without prior written permission of the copyright holder except as provided below

1. Any material contained in or derived from this unpublished research may be used by others in their writing with due acknowledgement.

2. IIUM or its library will have the right to make and transmit copies (print or electronic) for institutional and academic purposes.

3. The IIUM library will have the right to make, store in a retrieved system and supply copies of this unpublished research if requested by other universities and research libraries.

By signing this form, I acknowledged that I have read and understand the IIUM Intellectual Property Right and Commercialization policy.

Affirmed by Muhammad Nazreen Bin Muhammad Nor

……..…………………….. ………………………..

Signature Date

ACKNOWLEDGEMENTS

Alhamdulillah, praise be to Allah subhanahu wa ta'ala, the Merciful, the Beneficial, for the blessing that inspires me in completing my thesis writing.

I wish to express my gratitude and special thanks to Professor Datuk Paduka Dr Ridzwan Hashim, my main supervisor and Assistant Professor Dr Ibrahim Abu Bakar (alternate supervisor) for their continuous support, advice and encouragement throughout this journey. A special appreciation to my mentor and co-supervisor, Professor Dr Muhammad Nor Omar for his courage and support in finishing this program.

My appreciation and thanks to those who provided their time, effort and support for accomplishment of this project including staff of Kulliyyah of Science (Br Abdul Halim Ihsan and Ahmad Muzammil Zuberdi) and Kulliyyah of Allied Health Sciences. To members of my supervisory committee, I wish to thank for their supports. For staff and friends that I have not mentioned, thank you for your advice, supports and helps.

Last but not least, my utmost pleasure I dedicate this work to my beloved parents Rosnizahaini Abdul Ghani, Muhammad Nor Omar and members of the family, who supported me untill the end. Thank you for your support and patience.

TABLE OF CONTENTS

Abstract	ii
Abstract in Arabic	iii
Approval	iv
Declaration	v
Copyright Page	vi
Acknowledgements	x
List of Tables	xi
List of Figures	xii
CHAPTER ONE: INTRODUCTION	1
1.1 Research Background	1
1.2 Problem Statement	3
1.3 Research Question	5
1.4 Research Objective	5
1.5 Research Hypothesis	6
1.6 Significant of The Study	6
1.7 Study Overview	7
CHAPTER TWO: LITERATURE REVIEW	9
2.1 Rare Earth Element	10
2.2 Lanthanides including Lanthanum	11
2.3 Risk of rare earth element to environment and heatlh	11
2.3.1 Health effects	13
2.4 Bioaccumulation and phytoremediation	15
2.5 Bioaccumulation of rare earth metals by aquatic plant	18
2.6 Bioaccumulation of rare earth metals by freshwater fish	20
2.7 Inductive-Coupled Plasma Mass Spectrometry (ICP-MS)	24
CHAPTER THREE: RESEARCH METHODOLOGY	25
3.1 Materials	25
3.2 Methodology	26
3.2.1 Sample Preparation	27
3.2.2 Acid-wash Cleaning Reagent	28
3.2.3 Experimental Setup	28
3.2.3.1 Aquponics Tank Setup	28
3.2.3.2 Preparation of treatment solution containing LaCl3	28
3.2.3.3 Cultivation of *P. stratiotes* in Artificial Rare Earth Element wastewater containing lanthanum (La)	29
3.2.3.4 Rearing of *Oreochromis niloticus* in Artificial Rare Earth element containing lanthanum (La)	30
3.2.3.5 Validation analysis using certified reference material (CRM)	30
3.2.3.6 Digestion of *P.stratiotes* plant part containing Lanthanum	31
3.2.3.7 Digestion of *O. niloticus* fish tissues containing Lanthanum	32
3.2.3.8 Analysis of Blank Sample	33
3.2.4 Quantitative Analysis of Lanthanum concentration using ICP-MS	34
3.2.4.1 Calibration Graph	34
3.2.4.2 Analysis of Accumulated Lanthanum in *P.stratiotes and* O. niloticus	34
3.2.4.3 Calculation for determination of Lanthanum concentration	34

3.2.5 Statistical Analysis 34

CHAPTER FOUR: RESULTS AND DISCUSSION 35
4.1 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* roots 35
4.1.1 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* roots when exposed to 1ppm of LaCl3 artificial wastewater 35
4.1.2 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* roots when exposed to 3ppm of LaCl3 artificial wastewater 37
4.1.3 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* roots when exposed to 5ppm of LaCl3 artificial wastewater 39
4.1.4 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* roots when exposed to 10ppm of LaCl3 artificial wastewater 40
4.1.5 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* roots when exposed to LaCl3 artificial wastewater at different concentrations 42
4.2 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* leaves 46
4.2.1 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* leaves when exposed to 1ppm of LaCl3 artificial wastewater 46
4.2.2 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* leaves when exposed to 3ppm of LaCl3 artificial wastewater 49
4.2.3 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* leaves when exposed to 5ppm of LaCl3 artificial wastewater 50
4.2.4 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* leaves when exposed to 10ppm of LaCl3 artificial wastewater 51
4.2.5 Bioaccumulation of Lanthanum (La) in *Pistia stratiotes* leaves when exposed to LaCl3 artificial wastewater at different concentrations 53
4.3 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* Stomach 58
4.3.1 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* stomach when exposed to 1ppm of LaCl3 artificial wastewater 58
4.3.2 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* stomach when exposed to 3ppm of LaCl3 artificial wastewater 60
4.3.3 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* stomach when exposed to 5ppm of LaCl3 artificial wastewater 61
4.3.4 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* stomach when exposed to 10ppm of LaCl3 artificial wastewater 62
4.3.5 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* stomach when exposed to LaCl3 artificial wastewater at different concentrations 64
4.4 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* flesh 66
4.4.1 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* flesh when exposed to 1ppm of LaCl3 artificial wastewater 66
4.4.2 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* flesh when exposed to 3ppm of LaCl3 artificial wastewater 68
4.4.3 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* flesh when exposed to 5ppm of LaCl3 artificial wastewater 69

4.4.4 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* flesh when exposed to 10ppm of LaCl3 artificial wastewater 70
4.4.5 Bioaccumulation of Lanthanum (La) in *Oreochromis niloticus* 73

flesh when exposed to LaCl3 artificial wastewater at different concentrations
4.4.6 Comparison of the accumulation of lanthanum (La) in *Oreochromis niloticus* stomachs and flesh exposed in the REE artificial wastewater at different concentrations 74
4.4.7 Comparison of the accumulation of lanthanum (La) in *P. stratiotes* roots and leaves exposed in the REE artificial wastewater at different concentrations 77

CHAPTER FIVE: CONCLUSION AND FUTURE RESEARCH 81
5.1 Conclusion 82
5.2 Future Research 82

REFERENCES 83

APPENDIX A 88
APPENDIX B 89
APPENDIX C 90
APPENDIX D 91
APPENDIX E 92
APPENDIX F 93

LIST OF TABLES

Table No.		Page No.
2.1	Classification of *Pistia stratiotes*	20
2.2	Classification of *Nile Tilapia*	21
4.1.1	The accumulation of La in *Pistia stratiotes* Roots exposed in REE artificial wastewater at different concentration	36
4.2.1	The accumulation of La in *Pistia stratiotes* leaves exposed in REE artificial wastewater at different concentration	48
4.3.1	The accumulation of La in *Oreochromis niloticus* stomach exposed in REE artificial wastewater at different concentration	58
4.4.1	The accumulation of La in *Oreochromis niloticus* lesh exposed in REE artificial wastewater at different concentration	67

LIST OF FIGURES

Figure No.		Page No.
3.1	Aquaponic tanks containing different concentration of Lanthanum	26
3.2	Flowchart of the research experiment	27
4.1.1	The accumulation of Lanthanum in *Pistia stratiotes* roots exposed in 1 ppm REE artificial wastewater	37
4.1.2	The accumulation of Lanthanum in *Pistia stratiotes* roots exposed in 3 ppm REE artificial wastewater	38
4.1.3	The accumulation of Lanthanum in *Pistia stratiotes* roots exposed in 5 ppm REE artificial wastewater	40
4.1.4	The accumulation of Lanthanum in *Pistia stratiotes* roots exposed in 10 ppm REE artificial wastewater	41
4.1.5	The accumulation of Lanthanum in *Pistia stratiotes* roots exposed in REE artificial wastewater at different concentrations.	43
4.2.1	The accumulation of Lanthanum in *Pistia stratiotes* leaves exposed in 1 ppm REE artificial wastewater	46
4.2.2	The accumulation of Lanthanum in *Pistia stratiotes* leaves exposed in 3 ppm REE artificial wastewater	48
4.2.3	The accumulation of Lanthanum in *Pistia stratiotes* leaves exposed in 5 ppm REE artificial wastewater	51
4.2.4	The accumulation of Lanthanum in *Pistia stratiotes* leaves exposed in 10 ppm REE artificial wastewater	52
4.2.5	The accumulation of Lanthanum in *Pistia stratiotes* leaves exposed in REE artificial wastewater at different concentrations.	53
4.3.1	The accumulation of Lanthanum in *Oreochromis niloticus* stomach exposed in 1 ppm REE artificial wastewater	59
4.3.2	The accumulation of Lanthanum in *Pistia stratiotes* and *O*reochromis *niloticus* stomach exposed in 3 ppm REE artificial wastewater	60
4.3.3	The accumulation of Lanthanum in *Pistia stratiotes* and *Oreochromis niloticus* stomach exposed in 5 ppm REE artificial wastewater	62

4.3.4 The accumulation of Lanthanum in *Pistia Stratiotes* and *Oreochromis niloticus* stomach exposed in 10 ppm REE artificial wastewater 63

4.3.5 The accumulation of Lanthanum in *Pistia stratiotes* and *Oreochromis niloticus* stomach exposed in REE artificial wastewater at different concentrations. 64

4.4.1 The accumulation of Lanthanum in *Pistia stratiotes* and *Oreochromis niloticus* flesh exposed in 1 ppm REE artificial wastewater 67

4.4.2 The accumulation of Lanthanum in *Pistia Stratiotes* and *Oreochromis niloticus* flesh exposed in 3 ppm REE artificial wastewater 68

4.4.3 The accumulation of Lanthanum in *Pistia sratiotes* and *Oreochromis niloticus* flesh exposed in 5 ppm REE artificial wastewater 70

4.4.4 The accumulation of Lanthanum in *Pistia stratiotes* and *Oreochromis niloticus* flesh exposed in 10 ppm REE artificial wastewater 72

4.4.5 The accumulation of Lanthanum in Pistia s*tratiotes* and *Oreochromis niloticus* flesh exposed in REE artificial wastewater at different concentrations. 73

4.4.6 The accumulation of Lanthanum (La) in *Oreochromis niloticus* stomach and flesh exposed in the REE artificial wastewater at different concentration 76

4.4.7 The accumulation of Lanthanum (La) in *Pistia stratiotes* roots and leaves exposed in the REE artificial wastewater at different concentration 78

CHAPTER ONE

INTRODUCTION

1.1 RESEARCH BACKGROUND

Rare earth elements (REEs) are a group of elements listed at the bottom of the periodic table, specifically the elements classified as lanthanides. These include elements such as lanthanum (La), cerium (Ce), praseodymium (Pr), neodymium (Nd), promethium (Pm), samarium (Sm), europium (Eu), gadolinium (Gd), terbium (Tb), dysprosium (Dy), holmium (Ho), erbium (Er), thulium (Tm), ytterbium (Yb), and lutetium (Lu). Lanthanides are further grouped into light rare earth elements and heavier rare earth elements (Humphries, 2015). While named rare earths, they are in fact not that rare and are relatively abundant in the earth's crust. Previously, the REEs were produced in the USA, but over the last 20 years they were imported from China due to inexpensive in cost of production. According to United States Environmental Protection Agency (2012) and Lynas Corporation Limited (2013), elements like lanthanum have been used in producing items like electric car batteries, high-end and green technology devices.

In China, where 95 percent of all rare earths worldwide are mined, a lack of environmental regulation has allowed massive surface of groundwater pollution. Among the environmental problems caused by the mining and refinement of strategic elements, in this case speaking primarily about rare earth elements, is the problem of waste management. This can result in soil and water contamination by substances such as heavy metals and radioactive materials. This affects the ecosystem around the waste disposal site; and, if the contaminants get into the water table, it can affect areas beyond the site. As reported by Hurst (2010), The Chinese Society of Rare Earths stated that every ton of rare earth produced generates approximately 13 kg of dust. Since there are wide applications of rare earth elements (REEs), particularly in Western countries, the demand is growing and crucial for a country's economic sector.

Due to the importance of REE, Malaysia has set up Lynas Advance Materials Plant (LAMP) in 2009 near Kuantan Pahang and was issued the 2-year Full Operating Stage License (FOSL) in September 2014 by the Atomic Energy Licensing Board (AELB) under the Ministry of Science, Technology and Innovation (MOSTI) (Australian Mining, 2014). However, during the production and refining of REEs, the activity is susceptible in causing environmental contamination due to the wastewater residue containing REE and radioactive traces (Al-Areqi *et al*., 2014). Under the Temporary Operating License (TOL) issued by AELB (MOSTI) in September 2012, Lynas agreed to remove the radioactive residue out of Malaysia (AELB 2012). The removal of radioactive waste should be taken greater attention as it becomes a focus agenda under 11th Malaysian Plan (Strategy B5 - Managing waste holistically) (11th Malaysia Plan, 2015). Besides the residue containing radioactive materials, the REEs wastewater also contains metals (heavy and rare earth metals), once it enters the environment, they are hard to be destroyed but can be changed into another form (Zhang *et al*., 2013). After entering into the environment, these harmful substances have a tendency to be accumulated by plants and animals (including fish). Until to date, very little efforts have been taken in monitoring toxic radioactive wastewater discharged by Lynas after extracting REEs.

However, it is postulated that the environmental problem related to the rare earth elements (REEs) and radioactive contamination can be solved by generating the environment-friendly measures called phytoremediation through bioaccumulation especially by using aquatic plants (Singh *et al.* 2012). This assumption is based on the recent reports which used aquatic plants as biological methods for removing heavy metal content through bioaccumulation. As an example, studies conducted on treatment of industrial wastewater site using aquatic plants (*Pistia stratiotes*, *Eichomia crassipes* and *Salvinia molesta*) and freshwater fish (*Tilapia* and *Catfish*) found that the heavy metal content such as Pb, Cr and Cd have been accumulated in aquatic plants and fish (Baharom and Ishak, 2015). While reports also found that the radioactive elements such as uranium and thorium have been accumulated in aquatic plants and fish (Favas and Pratas, 2013; Labrot *et al*., 1999).

Several studies showed that the REEs could be accumulated in freshwater fish and plant system as conducted in examination of rare earth element concentration patterns in freshwater fish tissues (Mayfield and Fairbrother, 2015). Based on previous data and due to increase applications of REEs which resulted in environmental contamination, there has been a growing interest on the study of bioaccumulation processes of REEs using aquatic plants and freshwater fish.

1.2 PROBLEM STATEMENT

The wide application of rare earth elements (REEs) indicates that the demand is growing and crucial for a country's economic sector. Due to the importance of REE industry towards realisation of Vision 2020, Malaysia has set up a REE refining plant by establishing Lynas Advance Materials Plant (LAMP) in 2009 near Kuantan Pahang and was issued the 2-year Full Operating Stage License (FOSL) in September 2014 by the Atomic Energy Licensing Board (AELB) under the Ministry of Science, Technology and Innovation (MOSTI) (Australian Mining, 2014). However, during the production and refining of REEs, the activity is susceptible in causing environmental contamination due to the wastewater residue containing REE and radioactive traces (Al-Areqi *et al*, 2014). Under the TOL operating licence issued by AELB (MOSTI) in September 2012, Lynas agreed to remove the radioactive residue out of Malaysia (AELB, 2012). The removal of radioactive waste should be taken greater attention as it becomes a focus agenda under 11th Malaysian Plan (Strategy B5 - Managing waste holistically) (11th Malaysia Plan, 2015). Besides the residue containing radioactive materials, the REEs wastewater also contains metals (heavy and rare earth metals), once it enters the environment, they are hard to be destroyed but can be changed into another form (Zhang *et al*., 2013).

After entering into the environment, these harmful substances have a tendency to be accumulated by plants and animals (including fish). Until to date, very little efforts have been taken in monitoring toxic radioactive wastewater discharged by Lynas after extracting REEs. However, it is postulated that the environmental problem related to the rare earth elements (REEs) and radioactive contamination can be solved by generating the environment-friendly measures called phytoremediation through bioaccumulation especially by using aquatic plants (Singh *et al*, 2012). This assumption is based on the recent reports which used aquatic plants as biological methods for removing heavy metal content through bioaccumulation. As an example, studies conducted on treatment of industrial wastewater site using aquatic plants (*Pistia stratiotes*, *Eichomia crassipes* and *Salvinia molesta*) and freshwater fish (tilapia and catfish) found that the heavy metal content such as Pb, Cr and Cd have been accumulated in aquatic plants and fish (Razak *et al*, 2013; Eneji *et al*., 2011; Low *et al*., 2015; Baharom and Ishak, 2015). While reports also found that the radioactive elements such as uranium and thorium have been accumulated in aquatic plants and fish (Favas and Pratas, 2013; Labrot *et al*., 1999). Lastly, several studies showed that the REEs could be accumulated in freshwater fish and plant system (Mayfield and Fairbrother, 2015; Brioschi *et al*, 2013). Based on previous data and due to increase applications of REEs which resulted in environmental contamination, there has been a growing interest on the study of bioaccumulation processes of REEs using aquatic plants and freshwater fish.

1.3 RESEARCH QUESTIONS

i. How much can the aquatic *Pistia stratiotes* and *Oreochromis niloticus* remove the rare earth element, lanthanum from the artificial REEs industrial wastewater?

ii. What are the concentration of rare earth element, lanthanum accumulated in *P. stratiotes* and *O. niloticus* after cultivating them in artificial REE industrial wastewater?

iii. How is the green technology of phytoremediation using aquatic plant and fish able to treat the artificial REE industrial wastewater containing lanthanum residue?

1.4 RESEARCH OBJECTIVE

The general objective was to determine how much *Pistia stratiotes* (aquatic plant) and *Oreochromis niloticus* (freshwater fish) accumulate rare earth elements (REEs).

The specific objectives were:

1. to analyse the system of REEs industrial wastewater using the aquatic plant (*Pistia stratiotes*) and freshwater fish (*O. niloticus*).
2. to compare between aquatic plant and freshwater fish which is more effective in

accumulating Lanthanum.

3. to compare the bioaccumulation trend of REEs in plant and freshwater fish.

1.5 RESEARCH HYPOTHESIS

Aquatic plants (especially *P. stratiotes*) and freshwater fish (*O. niloticus*) are capable of acquiring large quantities of trace elements and heavy metals (including rare earth elements). The aquatic plants and freshwater fish will act as biological phytoremediation agents in removing and monitoring heavy metal and REEs residue content through green technology of bioaccumulation.

1.6 SIGNIFICANT OF THE STUDY

The major outcome of this study is the utilization of selected aquatic water lettuce plant (*Pistia stratiotes*) in removing REEs from industrial wastewater at refining plant. The result obtained will determine the reliability of using aquatic plants in removing REEs contamination. The utilization of aquatic plants in removing REEs toxic wastes contribute to the green and sustainable chemistry since it uses natural aquatic plants in reducing and eliminating the production of hazardous chemicals (REEs) that can potentially damage the environment.

The secondary outcome is that, after treating the REEs industrial wastewater, the resultant effluent discharged into the environment would be free from REEs toxic and radioactive materials and harmless to other aquatic life, including fish. This can be achieved by assessing the level of REEs in tilapia (*Oreochromis niloticus*). The REEs refining plant, for example Lynas Corporation Malaysia will benefit from this project to comply with the government regulation of toxic waste disposal procedures.

In addition, the treatment cost would be minimized. Finally, the community near to Lynas refining plant will not be affected as the resultant effluent discharged from industrial wastewater would be free from toxic radioactive REEs waste. The fish and plants harvested at nearby ponds and rivers will be safe for their consumption.The successful utilization of aquatic plants in controlling and removing toxic REEs waste will ensure the resultant discharged effluents from REEs wastewater pond into the environment are safe and would be harmless to human and aquatic life (*Tilapia* fish). Since the aquatic plants are abundantly available, the treatment of toxic REEs using these plants would not be expensive. Besides that, the plants are easily bred and grown in any aquatic condition, thus their utilizations in removing REEs waste could be scalable and replicable.

1.7 STUDY OVERVIEW

The research was conducted in stages while the writing of the thesis was divided into five chapters. As usual for a research which only deals with one variable, i.e. the concentration of lanthanum in aquatic plant and freshwater fish, Chapter 1 and 2 were specified for the Introduction and Literature Review respectively. In Introduction (Chapter One), a brief description of the rare earth elements (REEs) were highlighted including the importance of the REEs in modern technology and finally the environmental problems facing by the REEs manufacturing. Explanations on research background, objective, hypothesis and research problem related to REEs were described in this chapter. Meanwhile, Chapter Two was specified for Literature Review which listed some of the previously reported works done on REEs including the bioaccumulation of REEs in *Pistia stratiotes* and *Oreochromis niloticus*. A detail explanation on how the aquatic plant (*P. stratiotes*) and freshwater fish (*O. niloticus*) were sampled, treated and digested were described in Chapter Three. After digesting, the total accumulated lanthanums were analyzed using ICP-MS. Finally, Chapter Four highlighted the Results and Discussion which covered all analysis done in Chapter Three. Last but not least, Chapter Five which concluded all data from previous chapters under Conclusion and Future Research.

CHAPTER TWO

LITERATURE REVIEW

2.1 RARE EARTH ELEMENT

Rare earths are a series of chemical elements found in the Earth's crust that are vital to many modern technologies, including consumer electronics, computers, communications and clean energy (Brioschi *et al*., 2013). It is also known as rare earth elements (REEs) with unique elements grouped in the periodic table whereby they are known as lanthanides series from lanthanum to lutetium, scandium and yttrium (Zhang *et al*., 2013). Seventeen rare earth elements (REEs) in the periodic table includes 15 chemical elements called lanthanides. These include elements such as lanthanum (La), cerium (Ce), praseodymium (Pr), neodymium (Nd), promethium (Pm), samarium (Sm), europium (Eu), gadolinium (Gd), terbium (Tb), dysprosium (Dy), holmium (Ho), erbium (Er), thulium (Tm), ytterbium (Yb), and lutetium (Lu) (Please refer to Periodic Table). Traditionally, the REEs are divided into two groups on the basis of atomic weight, i.e. the light REEs are lanthanum through gadolinium (atomic numbers 57 through 64); and the heavy REEs comprise terbium through lutetium (atomic numbers 65 through 71). These lanthanides are further classified into light rare earth elements and heavier rare earth elements. They were named as "rare" because they were obtained from uncommon minerals in the 18^{th} and 19^{th} centuries. However, in the 21st century, the REEs have gained visibility through many media outlets because the public has recognized the critical, specialized properties that REEs contribute to modern technology, as well as China's dominance in production and supply of the REEs and (3) international dependence on China for the majority of the world's REE supply. According to Taylor and McClennan (1985), many REEs are in higher abundance compared to tin and molybdenum. China has been playing the role as the world's major producer of rare earth. It has provided 85– 95 percent of the world's REEs ((Long *et al*., 2010; Tse, 2011; Hatch, 2012). The REE usage increased substantially in high technology devices, including smart phones, digital cameras, computer hard disks, fluorescent and light-emitting-diode (LED) lights, flat screen televisions, computer monitors, and electronic displays. Large quantities of some REEs are used in clean energy and defense technologies (Goonan, 2011; Long, 2011; Tse, 2011; Hatch, 2012). However, in fulfilling global demands, aggressive production from rare earth mining could cause negative impacts towards the environment. In industrial areas, the wastewater due to the activities carried out may cause harmful effects to inhabitants surrounding it. The wastewater which contains the elements such as heavy metals must be treated before it is released into the environment. According to a commentary published by the Chinese Society of Rare Earths, "Every ton of rare earth produced, generates approximately 8.5 kilograms (18.7 lbs) of fluorine and 13 kilograms (28.7 lbs) of dust; and using concentrated sulfuric acid high temperature calcination techniques to produce approximately one ton of calcined rare earth ore generates 9,600 to 12,000 cubic meters (339,021 to 423,776 cubic feet) of waste gas containing dust concentrate, hydrofluoric acid, sulfur dioxide, and sulfuric acid, approximately 75 cubic meters (2,649 cubic feet) of acidic wastewater, and about one ton of radioactive waste residue (containing water)."

2.2 LANTHANIDES INCLUDING LANTHANUM

The lanthanides or lanthanoid series is a group of transition metals located on the periodic table in the first row (period) below the main body of the table. The lanthanides are commonly referred to as the rare earths, although many people group scandium and yttrium together with the rare earth elements. It's less confusing to call the lanthanides a subset of the rare earth metals.

2.3 RISK OF REE TO ENVIRONMENT AND HEALTH.

Human health risks associated with mining and manufacturing are typically associated either with contamination of potential sources of drinking water or ingestion of fine particulate mine waste (tailings). The human health effects of REEs have been sparsely studied since the work of Haley (1965). Ingestion and inhalation studies on cerium oxide and cerium compounds indicate minimal effects in laboratory studies on rats (U.S. Environmental Protection Agency, 2009). Exposure to cerium oxides and cerium compounds is unlikely during routine mining, although the potential for exposure is present during ore processing and refining. The United States lacks drinking water standards for the REEs. Additionally, the REE separation and refining process known as saponification had been used extensively in China until recently, generating harmful wastewater. It was estimated that, in 2015, the process generated 20,000 to 25,000 tons of wastewater, with total ammonia nitrogen concentrations ranging between 300 mg/L and 5,000 mg/L. The limited data on the toxicity of REEs in drinking water has been reviewed (de Boer *et al*., 1996). Information relevant to the environmental aspects of REE manufacturing is limited. Little is known about the aquatic toxicity of REEs. The concentrations of REEs in environmental ecosystems are influenced by their low abundances in crustal rocks and their limited solubility in