

**GENETIC IDENTIFICATION OF SELECTED ORNAMENTAL
FISHES IN SEREMBAN, NEGERI SEMBILAN**

WAN FARA ASYIKIN WAN ZAINAL AZHAR

**BACHELOR OF SCIENCE (Hons.) BIOLOGY
FACULTY OF APPLIED SCIENCE
UNIVERSITI TEKNOLOGI MARA**

JANUARY 2019

The Final Year Project Report entitled “**Genetic Identification of Selected Ornamental Fishes in Seremban, Negeri Sembilan**” was submitted by Wan Fara Asyikin Wan Zainal Azhar, in partial fulfillment of the requirements for the Degree of Bachelor of Science (Hons.) Biology, in the Faculty of Applied Sciences, and was approved by

Dr. Izzati Adilah binti Azmir
Supervisor
Faculty of Applied Sciences
Universiti Teknologi Mara (UiTM)
Negeri Sembilan, Kampus Kuala Pilah,
Pekan Parit Tinggi, 72000 Kuala Pilah,
Negeri Sembilan

Siti NorAzura binti Jamal
Coordinator FSG661 AS201
Faculty of Applied Sciences
Universiti Teknologi Mara (UiTM)
Negeri Sembilan, Kampus Kuala Pilah,
Pekan Parit Tinggi, 72000 Kuala Pilah,
Negeri Sembilan

Dr. Aslizah binti Mohd Aris
Head of Biology School
Faculty of Applied Sciences
Universiti Teknologi Mara (UiTM)
Negeri Sembilan, Kampus Kuala Pilah,
Pekan Parit Tinggi, 72000 Kuala Pilah,
Negeri Sembilan

Date: _____

TABLE OF CONTENTS

	PAGE
ACKNOWLEDGEMENT	iii
TABLE OF CONTENTS	iv
LIST OF TABLES	vi
LIST OF FIGURES	vii
LIST OF ABBREVIATIONS	viii
ABSTRACT	ix
ABSTRAK	x
CHAPTER 1: INTRODUCTION	
1.1 Background of Study	1
1.2 Problem Statement	2
1.3 Significance of Study	3
1.4 Objectives of Study	3
CHAPTER 2: LITERATURE REVIEW	
2.1 Ornamental fishes in Malaysia	4
2.1.1 <i>Puntigrus tetrazona</i>	6
2.1.2 <i>Balantiocheilos melanopterus</i>	7
2.1.3 <i>Carrasius auratus</i>	8
2.1.4 <i>Betta splendens</i>	9
2.1.5 <i>Xiphophorus helleri</i>	10
2.1.6 <i>Pterophyllum altum</i>	11
2.1.7 <i>Poecilia sphenops</i>	12
2.2 Socio-economic importance of ornamental fishes	12
2.3 Ornamental freshwater fish trade	14
2.4 DNA barcoding	16
2.5 The promise of DNA barcoding	17
CHAPTER 3: METHODOLOGY	
3.1 Materials	
3.1.1 Raw materials	19
3.1.2 Chemicals	20
3.1.3 Apparatus	21
3.2 Methods	
3.2.1 Samples collection	21
3.2.2 Genomic DNA extraction	23
3.2.3 Quantification and quality determination of DNA	24
3.2.4 PCR amplification and agarose gel electrophoresis	25
3.2.5 DNA sequencing	27

3.3	Data Analysis	
3.3.1	Sequences submission to BOLD system	28
3.3.2	CLUSTAL X	28
3.3.3	BIOEDIT	28
3.3.4	MEGA 7.0	29
	CHAPTER 4: RESULTS AND DISCUSSION	
4.1	Amplification of the PCR products	30
4.2	Species identification by pre-existing sequences in the database	32
4.3	DNA sequences assignment	33
4.4	Ornamental fish <i>COI</i> gene assignment	34
4.5	Phylogenetic relationship between ornamental fishes	37
	CHAPTER 5: CONCLUSIONS AND RECOMMENDATIONS	41
	CITED REFERENCES	42
	APPENDICES	51
	CURRICULUM VITAE	52

ABSTRACT

GENETIC IDENTIFICATION OF SELECTED ORNAMENTAL FISHES IN SEREMBAN, NEGERI SEMBILAN

The ornamental fish sector is a widespread and global component of international trade, fisheries, aquaculture and development. The utilization of multiple trade names causes problem in species identification. Moreover, unmanaged trading could lead to severe threats to biodiversity. In this regard, DNA barcoding could effectively clarify the divergence of the species. Considering the utility of DNA barcoding as a comprehensive system for species identification and discovery, this study aims to investigate the genetic relationship and to construct the phylogenetic tree among those selected fish species collected from selected pet stores in Seremban, Negeri Sembilan. The 642bp barcode fragment of the Cytochrome *c* oxidase I (*COI*) gene was PCR amplified. Results from BLAST showed all the generated sequence were subjected to high percentage identity index and similarity between 99% to 100%. It was then analyzed using MEGA 7.0 through Neighbour-Joining (NJ) clustering and K2P distance based approach. The analysis revealed straightforward identification of eight specimens into five species with increasing value of genetic distances from conspecific (0.05%) to the taxonomic level (20.18%). The phylogenetic analysis consists of own sequences and reference sequences obtained from the GenBank. All the specimens from different genus was found with high bootstrap value ($n > 90\%$) through Neighbour-Joining (NJ) and Maximum Likelihood method. Thus, DNA barcoding reflects the efficacy of the techniques in identifying the genetic assessment in selected ornamental fishes.