GENETIC VARIATION OF MALAYSIAN GOLD AROWANA (*Scleropages formosus*) USING PCR-RFLP ON CYTOCHROME B GENE

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UNIVERSITY MALAYSIA TERENGGANU
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By

SUGENDRA KUMAR A/L VINODAMANEY

Research Report submitted in partial fulfillment of

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SCHOOL OF MARINE SCIENCE AND ENVIRONMENT
UNIVERSITI MALAYSIA TERENGGANU

DECLARATION AND VERIFICATION REPORT

FINAL YEAR RESEARCH PROJECT

It is hereby declared and verified that this research report entitled Genetic Variation of Malaysian Gold Arowana (*Scleropages formosus*) Using PCR-RFLP on Cytochrome B Gene **by Sugendra Kumar A/L Vinodamaney**, Matric No. **UK25295** have been examined and all errors identified have been corrected. This report is submitted to the School of Marine Science and Environment as partial fulfillment towards obtaining the **Degree of Science (Marine Biology)**, School of Marine Science and Environment, Universiti Malaysia Terengganu.

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LIST OF ABBREVIATIONS

%	Percentage
° C	Degree celsius
µl	Microlitre
Bp	Base pairs
Cm	Centimeters
DNA	Deoxyribonucleic Acid
dNTP	Deoxynucleotide triphosphate
G	Gram
$MgCl_2$	Magnesium chloride
Ml	Milliliter
ng / µl	Nanogram per Microlitre
PCR	Polymerase Chain Reaction
RFLP	Restriction fragment Length polymorphism
Rpm	Rotation per minute
V	Voltage

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ABSTRACTS

A study of genetic variation of Malaysian Gold Arowana or known as *Scleropages formosus* using PCR-RFLP method on cytochrome b gene. The project was done by using the fish scales which were taken from Bukit Merah, Perak. The aim of this study is to determine the genetic variation of *Scleropages formosus* using different types of restriction enzymes those are *Hae III, Mbo I, Nde I, Alu I* and *Msp I.* DNA was extracted and was amplified because for RFLP method large quantity of DNA is needed, the DNA was amplified by using Cytochrome b primer. After the amplification of DNA was done, purification of the PCR sample was done to remove the impurities which might affect the result of the study. Final procedure was the RFLP method by using Restriction enzymes, *Hae III, Mbo I, Nde I, Alu I* and *Msp I.* the DNA base pairs in PCR was about 1100 bp and this was continued with RFLP method. All the restriction enzymes have the specific recognition site to cut and this would form fragment during the agarose gel electrophoresis. After the PCR-RFLP method it was known that these enzymes cannot be used for determine the genetic variations among *Scleropages formosus* as the cutting sites and the base pairs were similar for all the samples of *Scleropages formosus* thus the differences among the species population could not be detected. So it is recommended to test the genetic variability among *Scleropages formosus* using different restriction enzymes.

Variasi Genetik Arowana Emas (*Scleropages formosus)* dengan Menggunakan Kaedah PCR-RFLP pada Gene Sitokrom b

ABSTRAK

Variasi dalam genetik ikan arowana emas (*Scleropages formosus)* dilakukan dengan mengunakan kaedah PCR-RFLP pada bahagian Cytochrome b gene. Pengenalan pelajaran ini adalah untuk menentukan variasi dalam genomik *Scleropages formosus* dengan mengunakan enzim sekatan yang dikenali sebagai *Hae III, Mbo I, Nde I, Msp I* dan *Alu I.* sampel ikan arowana iaitu sisik dan sisik arowana telah didapatkan dari Bukit Merah, Perak tempat hatcheri arowana yang utama. DNA dari sisik ikan kelisa telah di ekstrak dan DNA ini telah dikembangkan dengan mengunakan primer yang dikenali sebagai primer Cytochrome b. tujuan utama amplifikasi DNA adalah untuk membanyakkan kepekatan DNA untuk kaedah yang seterusnya RFLP. RFLP telah dilakukan dengan menggunakan 5 jenis Enzim pemotong yang telah dinyatakan. Keputusan elektroforesis menunjukkan bahawa enzim pemotong ini tidak sesuai untuk digunakan untuk menentukan variasi dalam ikan kelisa emas disebabkan tidak perbezaan yang dikesan pada cebisan DNA yang telah dipotong untuk kesemua sampil. Manakala enzim pemotong *Msp I* dan *Alu I* pula tidak potong sampil DNA ini disebabkan urutan pengenalan enzim ini adalah berlainan daripada sampil. Penggunaan enzim pemotong lain untuk mencuba mengesan variasi dalam ikan kelisa adalah digalakkan.