

RESEARCH ON THE LIFE HISTORY OF *CASSIDULA*  
*ATRISETTUS* AND ITS BIOLOGICAL CONTROL  
BY *PHANOCERA* AND *TRICHOPTERUS*  
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Study on genetic variability of *Cassidula aurisfelis* (snail) by using random amplified polymorphic dna (rapd) polymerase chain reaction (pcr) technique / Mohammad Shafie Shafie.

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**STUDY ON GENETIC VARIABILITY OF *CASSIDULA AURISFELIS* (SNAIL)  
BY USING RANDOM AMPLIFIED POLYMORPHIC DNA (RAPD) -  
POLYMERASE CHAIN REACTION (PCR) TECHNIQUE**

**By**

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**Research Report submitted in partial fulfillment of  
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**JABATAN SAINS BIOLOGI  
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**PENGAKUAN DAN PENGESAHAN LAPORAN  
PROJEK PENYELIDIKAN I DAN II**

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: STUDY ON GENETIC VARIABILITY OF *CASSIDULA AURISFELIS* (SNAIL) BY USING RANDOM AMPLIFIED POLYMORPHIC DNA (RAPD) - POLYMERASE CHAIN REACTION (PCR) TECHNIQUE, oleh Mohammad Shafie Bin Shafie, no. matrik: UK 7934 telah diperiksa dan semua pembetulan yang disarankan telah dilakukan. Laporan ini dikemukakan kepada Jabatan Sains Biologi sebagai memenuhi sebahagian daripada keperluan memperoleh Ijazah Sarjana Muda Sains (Sains Biologi), Fakulti Sains dan Teknologi, Kolej Universiti Sains dan Teknologi Malaysia.

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## LIST OF ABBREVIATIONS

%	Percentage
°C	Degree Celsius
1X	One Time
A	Adenosine
bp	Base pair
C	Cytosine
cm	Centimeter
dH <sub>2</sub> O	Distilled water
DNA	Deoxyribonucleic acid
dNTP mix	Deoxyribonucleotides mixture
EDTA	Ethylenediaminetetracetic acid
g	Gram
G	Guanocine
M	Molarity
µg	Microgram
µL	Microlitre
µM	Micromolar
mg	Miligram
mL	Mililitre
mM	Milimolar
min	Minutes
ng	Nanogram
OD	Optical density

PCR	Polymerase Chain Reaction
Pmole	Picomole
Ppt	Part per trillion
RAPD	Random Amplified Polymorphic DNA
rpm	Rotation per minute
sec	Seconds
SD	Standard Deviation
SDS	Sodium Dodecyl Sulfate
T	Thymine
TBE	Tris-borate-EDTA buffer
TE	10mM Tris Cl, 1 mM EDTA
TNES	Tris NaCl EDTA-2Na SDS
Tris-HCL	Tris [Hydroxymethyl] aminomethane hydrochloride
UV	Ultra violet
V	Volt
VDS	Video Documentation System
v/v	volume/volume
w/v	weight/volume

## ABSTRACT

The genetic variability among individuals of *Cassidula aurisfelis* from Setiu Wetland, Terengganu Darul Iman was examined by using the random amplified polymorphic DNA (RAPD) technique. The genomic DNA was extracted from the samples tissues using Kit Wizard™ Genomic DNA Purification (Promega) and Phenol-chloroform method. Phenol-chloroform method was selected to use for the next step. The results produced by the machine showed clear RAPD banding pattern. Ten oligonucleotide primers (Kit A) were screened and three primers were selected (OPA 02, OPA 04 and OPA 10) to amplify DNA from five samples of *Cassidula aurisfelis* from Setiu Wetland, Terengganu. A total of 28 RAPD fragments (RAPDs) with 17 polymorphic fragments (60.71%) with size ranging from 300 – 1750 bp were scored from the population. Genetic distance for samples ranges from 0.135 to 0.269. For similarity index samples ranges from 0.7179 to 0.8649 (mean  $0.7810 \pm 0.0497$ ).

**KAJIAN MENGENAI KEPELBAGAIAN GENETIK *CASSIDULA AURISFELIS* (SIPUT) DENGAN MENGGUNAKAN TEKNIK AMPLIFIKASI RAWAK DNA POLIMORFIK (RAPD) - TINDAKBALAS RANTAI POLIMERASE (PCR)**

**ABSTRAK**

Kepelbagaian genetik di antara individu-individu *Cassidula aurisfelis* dari Setiu Wetland, Terengganu Darul Iman, telah dikaji dengan menggunakan kaedah Amplifikasi Rawak DNA Polimorfik (RAPD). Genomik DNA telah diekstrak daripada tisu dengan menggunakan ‘Kit Wizard™ Genomic DNA Purification (Promega)’ dan ‘Phenol-chloroform method’. ‘Phenol-chloroform method’ telah dipilih untuk langkah seterusnya. Hasil keputusan diperolehi daripada mesin menunjukkan jalur – jalur RAPD yang jelas. Sepuluh pencetus oligonukleotida (Kit A) telah diuji dan tiga daripada pencetus (OPA 02, OPA 04 dan OPA 10) telah dipilih untuk mengamplifikasikan DNA daripada lima sampel yang dipilih daripada Setiu Wetland, Terengganu. Sejumlah 28 jalur RAPD dan 17 jalur RAPD yang polimorfik (60.71%) yang bersaiz diantara 300 – 1750 bp telah dihasilkan dan dikenalpasti. Paras jarak perbezaan genetik antara sampel adalah daripada 0.135 hingga 0.269. Untuk indek persamaan antara sampel adalah daripada 0.7179 sehingga 0.8649 (purata  $0.7810 \pm 0.0497$ ).