

INDUCTION AND ESTABLISHMENT OF IN VITRO
CULTURE OF *Homalium malayanum*

MOHD NUR SIDDIQ AHMAD TALIB

FACULTY SAINS DAN TEKNOLOGI
KOLEJ UNIVERSITI SAINS DAN TEKNOLOGI MALAYSIA
2005

01/2054

1100036815

LP 22 FST 1 2005



1100036815

Induction and establishment of in vitro culture of hanguana
malayana / Mohd Nasir Ahmad Tajudin.



PERPUSTAKAAN
KOLEJ UNIVERSITI SAINS & TEKNOLOGI MALAYSIA
21030 KUALA TERENGGANU

1100036815		

Lihat sebelah

HAK MILIK
PERPUSTAKAAN KUSTEM

INDUCTION AND ESTABLISHMENT OF IN VITRO CULTURE OF
Hanguana malayana.

By

Mohd Nasir bin Ahmad Tajudin

Research Report submitted in partial fulfillment of
the requirements for the degree of
Bachelor Of Science (Biological Sciences)

Department of Biological Sciences
Faculty of Science and Technology
KOLEJ UNIVERSITI SAINS DAN TEKNOLOGI MALAYSIA
2005

This project should be cited as:

Nasir, A.T. 2004. Induction and establishment of in vitro culture of *Hanguana malayana*. Undergraduate thesis, Bachelor of Science in Biological Sciences, Faculty of Science and Technology, Kolej Universiti Sains dan Technology Malaysia. 27p.

No part of this project report may be produced by any mechanical, photographic, or electronic process, or in form of phonographic recording, nor may it be stored in a retrieval system, transmitted, or otherwise copied for public or private use, without written permission from the author and the supervisor(s) of the project.

KOLEJ UNIVERSITI SAINS DAN TEKNOLOGI MALAYSIA

**PENGAKUAN DAN PENGESAHAN LAPORAN
PROJEK PENYELIDIKAN I DAN II**

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: **Induction and establishment of in vitro culture of *Hanguana malayana*** oleh **Mohd Nasir Bin Ahmad Tajudin** No. Matrik Uk **6624** 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.

Disahkan oleh:



Penyelia Utama
Nama: **DR. AZIZ BIN AHMAD (Ph.D)**
LECTURER
Dept of Biological Sciences
Cop Rasmi: Faculty of Science and Technology
University College of Science
and Technology Malaysia
21030 Kuala Terengganu.

Tarikh: 6/4/2005

Penyelia Kedua (jika ada)

Nama:

Cop Rasmi

Tarikh:



PROF. MADYA DR. NAKISAH BT. MAT AMIN
Ketua Jabatan Sains Biologi
Nama: **Jabatan Sains Biologi**
Fakulti Sains dan Teknologi
Kolej Universiti Sains dan Teknologi Malaysia
Cop Rasmi: **(KUSTEM)**
21030 Kuala Terengganu.

Tarikh: 6/4/05

ACKNOWLEDGEMENT

First of all, thank god cause provide me with a lot of idea, to guide me with your bless until I got to complete my project with all of convenience. I hope this project on *Hanguana malayana* will be a pioneer to break the mystery that hiding behind this plants, and off course the breaking of puzzle will give a huge of benefit and a wealth of profit to the other peoples.

I would like to thank Dr Aziz bin Ahmad for helping and supervising me along the way to complete my final year project. His advices and his concerns have given me a spectrum of light; illuminate my direction to run my project accurately in order to achieve the objective given.

My appreciation also due to Abang Mazrul, Cik Azlina, Abas, and Kak Rokiah for their dedication to help me and their contribution in completing my project hardly could not be denied. For the names that are not being stated here, believe me, your name are always emerging all the time in the special place that is in my heart.

Last but not least, last but the strength of my mind, the support of my soul, and the courage of my attempt, to my family, my mum and my father, to

the special person, without you, the results will be nothing, but just only like a body without a soul.

TABLE OF CONTENTS

	Page
ACKNOWLEDGEMENT	ii
LIST OF TABLES	iii
LIST OF PLATES	iv
LIST OF SYMBOLS	v
ABSTRACT	vi
ABSTRAK	vii
1.0 INTRODUCTION	1
2.0 LITERATURE REVIEW	3
2.1 The <i>Hanguana malayana</i>	3
2.2 The problems	4
2.3 The previously application of tissue culture on aquatic plants	4
2.4 Culture media	6
3.0 MATERIALS AND METHODS	7
3.1 Sources of explants	7
3.2 Sterilization of explants	8
3.3 Medium for culture establishment	9

4.0	RESULTS	10
4.1	Sterilization of shoots culture	10
4.1.1	Shoot tips	10
4.1.2	Adventitious shoots	11
4.2	Establishment of shoot cultures	13
4.2.1	Shoot tips	13
4.2.2	Adventitious shoots	14
5.0	DISCUSSION	16
6.0	CONCLUSION	22
	REFERENCES	23
	VITAE CURICULUM	26

LIST OF TABLES

Tables		Page
1	Plants have been propagated using tissue culture technique	10
2	The percentage of explants free of contamination after treatment with various concentration of Clorox at different time of immersion.	13
3	The percentage of shoot tips free of contaminant after treatment with various concentration of Clorox at different times of immersion.	14
4	The percentage of shoot tips free of contaminant after treatment with various concentration of Clorox at different times of immersion with addition of 1g/l of activated charcoal, 100 mg/l of ascorbic acid and 100 mg/l of gentamicin sulphate.	16
5	The percentage of adventitious shoots free of contaminant after treatment with various concentrations of Clorox at different times of immersion.	17
6	The percentage of adventitious shoots free of contaminant after treatment with various concentration of Clorox at different times of immersion with addition of 1g/l of activated charcoal, 100 mg/l of ascorbic acid and 100 mg/l of gentamicin sulphate.	19
7	Describing the problems occurred for each samples immersed in different percentage of Clorox.	22

LIST OF PLATES

Plates	Page
1 The clump of <i>Hanguana malayana</i> from the sampling site located behind the domestic airport at Kuala Terengganu.	9
2 (a), (b) The culture contaminated by fungus and bacteria; (c), (d) healthy shoot tips and adventitious shoots; (e) from left to right, the explants on the test tube were contaminated by bacteria, the occurring of tissue browning and yeast.	20

LIST OF SYMBOLS

BAP	-	Benzylaminopurine
HCL	-	Hydrochloric acid
H ₂ O ₂	-	Hydrogen peroxide
Kin	-	Kinetin
L-AA	-	L-Ascorbic acid
M	-	Molar
mg/l	-	Milligram per liter
NaOH	-	Sodium hydrochloride
O ₂ ⁻	-	Superoxide
OH·	-	Hydroxyl radical
ROS	-	Reactive oxygen species

ABSTRACT

Attempt was made to establish on in vitro culture of *Hanguana malayana*. The surface sterilization practices were successfully obtained by using 30% (v/v) of Clorox with 30 minutes of immersion. Initiation of shoot cultures was established by addition of 1g/l of activated charcoal into media, 100 mg/l of L-ascorbic acid and 100 mg/l of gentamicin sulphate has been treated to the explants body. Two types of explants, adventitious shoots and shooting tips were used. The adventitious shoot was most successful. The percentage of no contaminant was 75% obtained from 250 of adventitious shoot cultured compared to only 57.14% by shooting tips. The problem interfering during the inoculation stage that could be observed was the high rate of contamination and the naturally occurring of oxidative stress.

ABSTRAK

Usaha untuk mendapatkan kultur in vitro pokok *Hanguana malayana* telah berjaya dilakukan. Kadar pensterilan yang optimum didapati dengan menggunakan 30% (v/v) klorox, direndam selama 20 minit. Permulaan bagi pertumbuhan kultur pokok pada masa kajian telah dilakukan dengan menambahkan 1 g/l arang teraktif ke dalam media, 100 mg/l L-ascorbic acid dan 100 mg/l gentamicin sulfat digunakan bagi merawat tumbuhan. Dua jenis ekplan telah digunakan iaitu tunas sisi dan tunas pucuk. Tunas sisi merupakan eksplant yang paling berjaya. Peratusan tanpa sebarang kontaminasi adalah 75% berbanding tunas pucuk yang hanya mencapai 57.14%. Masalah yang sering timbul ketika pengkulturan tisu tumbuhan ini ialah kadar kontaminasi yang tinggi dan tekanan oksidatif yang dialami oleh tisu yang dikulturkan.