

ISOLATION AND IDENTIFICATION OF FUNGI ASSOCIATED
WITH LEAVES OF CULTIVATED AND WILD PLANTS

BY R. S. CHAUHAN AND M. K. SHARMA

PHYSICO-MICROBIAL LABORATORY

DEPARTMENT OF BIOTECHNOLOGY
UNIVERSITY OF JAMMU & KASHMIR

2007

978 4290

1100051119 Perpustakaan Sultanah Nur Zahirah (UMT)
Universiti Malaysia Terengganu



LP 6 FST 2 2007



1100051119

Isolation and identification of fungi associated with Nypa
fruticans in Universiti Malaysia Terengganu, Terengganu /
Aznoorlina Zainuddin.

PERPUSTAKAAN

UNIVERSITI MALAYSIA TERENGGANU (UMT)

21030 KUALA TERENGGANU

1100051119

Lihat sebelah

HAK MILIK
PERPUSTAKAAN UMT

ISOLATION AND IDENTIFICATION OF FUNGI ASSOCIATED WITH *Nypa fruticans* IN UNIVERSITI MALAYSIA TERENGGANU, TERENGGANU

By

Aznoorlina binti Zainuddin

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
UNIVERSITY MALAYSIA TERENGGANU
2007

1100051119

This project should be cited as:

Aznoorlina, Z. 2007. Isolation and identification of fungi associated with *Nypa fruticans* in UMT, Terengganu. Undergraduate Thesis, Bachelor of Science (Biological Sciences), Faculty of Science and Technology, Universiti Malaysia Terengganu. 58p.

No part of this project report may be produced by any mechanical, photographic, or electronic process, or in the 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.



JABATAN SAINS BIOLOGI
FAKULTI SAINS DAN TEKNOLOGI
UNIVERSITI MALAYSIA TERENGGANU

PENGAKUAN DAN PENGESAHAN LAPORAN
PROJEK PENYELIDIKAN I DAN II
RESEARCH REPORT VERIFICATION

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: Isolation and Identification of Fungi Associated with *Nypa fruticans* in Universiti Malaysia Terengganu, Terengganu, oleh Aznoorlina Binti Zainuddin, no. matrik: UK10385 telah diperiksa dan semua pembetulan yang disarankan telah dilakukan. Laporan ini dikemukakan kepada Jabatan Sains Biologi sebagai memenuhi sebahagian daripada keperluan memperolehi ijazah Sarjana Muda Sains (Sains Biologi), Fakulti Sains dan Teknologi, Universiti Malaysia Terengganu.

Disahkan oleh: /Verified by:

Penyelia Utama/Main Supervisor
DR. MARIAM TAIB
Nama: Pensyarah
Cop Rasmi: Jabatan Sains Biologi
Fakulti Sains dan Teknologi
Universiti Malaysia Terengganu
21030 Kuala Terengganu.

Tarikh: 14/5/07

Penyelia Kedua (jika ada)/Co-Supervisor (if applicable)
JAMILAH MOHD SALIM @ HALIM
Nama: Pensyarah
Cop Rasmi: Jabatan Sains Biologi
Fakulti Sains dan Teknologi
Universiti Malaysia Terengganu
21030 Kuala Terengganu.

Tarikh: 14/5/07

Ketua Jabatan Sains Biologi/Head, Department of Biological Sciences

Nama: **DR. AZIZ BIN AHMAD**
Cop Rasmi: Ketua
Jabatan Sains Biologi
Fakulti Sains dan Teknologi
Universiti Malaysia Terengganu
21030 Kuala Terengganu

Tarikh: 14/5/07

ACKNOWLEDGEMENTS

In the name of Allah S.W.T The Most Beneficent and The Most Merciful. The deepest sense of gratitude to the Almighty for the strength and ability to complete this project.

I would like to take this opportunity to express my gratitude towards my supervisor Dr. Mariam Taib, my co-supervisor Miss Jamilah Mohd. Salim @ Halim, Microbiology Lab Assistants Mdm. Zarina and Kak Ti, and not to forget Dr. Cha Thye San as my Personal Advisor for their guidance, advice, and understanding during my final year project. Infinite thanks I brace upon them.

I would also like to express my deepest appreciation to my colleagues, lecturers, friends especially to Cik Mohd Nurani Mustaffa from PPUM for his help in printing this report, En Kamri for the support to encourage me to finish up this project and also to Cik Dzainor Shahril who is kind enough to lend me his laptop until I finished this thesis.

Also thanks and appreciation to my project group members; Tikah, Siti Ropiah, Jamal, Shu and Pit Li for their help during my lab work. Not forgetting my beloved friends Siti Nurul 'Ashikin, Fadilah, Shikin, Radiah, Eyoh, Raymond, Nik, Shima, and Ikin for the willingness on helping me in various ways no matter day or night with all the difficulties I faced.

Last but not least, I would like to express my deepest appreciation to my family members especially to both my parents Pn Aminah binti Moksin and En Zainuddin bin Abas and also to my sister Aznilinda, Azniza and Aznoorlita for their support physically, mentally, and spiritually.

Finally to individuals who has involved neither directly nor indirectly in succession of this thesis. Without them, this project cannot be successfully completed.

TABLE OF CONTENTS

	Page
ACKNOWLEDGEMENTS	ii
LIST OF TABLES	v
LIST OF FIGURES	vi
LIST OF ABBREVIATIONS	vii
LIST OF APPENDICES	viii
ABSTRACT	ix
ABSTRAK	x
CHAPTER 1 INTRODUCTION	1
1.1 Introduction	1
1.2 Importance of Study	4
1.3 Objectives of Study	4
 CHAPTER 2 LITERATURE REVIEW	5
2.1 Mangroves	5
2.2 The Importance of Mangroves	7
2.2.1 Economic Importance	8
2.2.2 Food Importance	10
2.2.3 Medicinal Importance	11
2.3 Tropical and Subtropical Mangroves	12
2.3.1 <i>Acrostichum aureum</i>	12
2.3.2 <i>Avicennia alba</i>	14
2.3.3 <i>Lumnitzera racemosa</i>	15
2.3.4 <i>Rhizophora apiculata</i>	16
2.3.5 <i>Sonneratia caseolaris</i>	17
2.3.6 <i>Nypa fruticans</i>	17
2.4 Microbes Associated With Mangroves	20
2.4.1 Marine Fungi	22
2.5 Bioactive Compounds of Fungi	22

CHAPTER 3 METHODOLOGY	26
3.1 Sampling	26
3.2 Isolation of Fungi Associated with <i>Nypa fruticans</i>	27
3.2.1 Direct Culture Technique	27
3.2.2 Damp Incubation Technique	28
3.3 Identification of Fungal Isolates	28
CHAPTER 4 RESULTS	30
4.1 Fungi isolated from <i>N. fruticans</i>	30
4.2 Fungi Isolated by Direct Culture Technique	31
4.2.1 Isolation of fungi from leaves	32
4.2.2 Isolation of fungi from fronds	35
4.2.3 Isolation of fungi from barks	36
4.3 Fungi Isolated by Damp Incubation Technique	38
4.3.1 Isolation of fungi from leaves	39
4.3.2 Isolation of fungi from fronds	41
4.3.3 Isolation of fungi from barks	42
CHAPTER 5 DISCUSSION	43
CHAPTER 6 CONCLUSION AND RECOMMENDATIONS	46
REFERENCES	47
APPENDICES	56
CURRICULUM VITAE	58

LIST OF TABLES

Table	Page
4.1 Summary of isolated fungi from <i>N. fruticans</i>	30
4.2 List of fungi isolated from <i>N. fruticans</i> using Direct Plating Technique	31
4.3 List of fungi isolated from <i>N. fruticans</i> using Damp Incubation Technique	38

LIST OF FIGURES

Figure		Page
2.1	<i>A. aureum</i> fern	13
2.2	<i>A. alba</i> tree	14
2.3	<i>A. alba</i> root with pencil-like pneumatophores	14
2.4	<i>L. racemosa</i> tree	16
2.5	<i>R. apiculata</i>	16
2.6	<i>S. caseolaris</i>	17
2.7	<i>N. fruticans</i> plant	18
2.8	Scientific classification of <i>N. fruticans</i>	18
3.1	Sampling site	26
3.2	Direct Plating Technique	29
3.3	Slide Culture Technique	29
3.4	Damp Incubation Technique	29
4.1	Fungi isolated from leaves in Direct Culture Technique	34
4.2	Fungi isolated from fronds in Direct Culture Technique	35
4.3	Fungi isolated from barks in Direct Culture Technique	36
4.4	Fungi isolated from leaves in Damp Incubation Technique	39
4.5	Fungi isolated from fronds in Damp Incubation Technique	42
4.6	Fungi isolated from barks in Damp Incubation Technique	42

LIST OF ABBREVIATIONS

\$	-	Dollar
%	-	Percent
'	-	Minutes
°	-	Degree
°C	-	Degree Celsius
cm	-	Centimeter
E	-	East
ft.	-	Feet
g	-	Gram
ha	-	Hectare
in.	-	Inch
Km ²	-	Kilometer squad
m	-	Meter
ml	-	Milliliter
mm	-	Millimeter
N	-	North
PDA	-	Potato Dextrose Agar
S	-	South
St.	-	State
SWA	-	Sea Water Agar
US	-	United State
μ	-	Micrometer

LIST OF APPENDICES

Appendix		Page
1	Preparation of media PDA and SWA	56
2	List of nutrients in preparing artificial sea water	57

ABSTRACT

Mangrove plants have a great potential in the production of bioactive compounds that can be used for medicinal purposes. However, it is not certain whether the bioactive compounds are produced by mangrove plant itself or by associated microbes. In order to determine this, fungi associated with *Nypa fruticans* were isolated. The sampling of fragments of leaves, fronds and barks of *N. fruticans* was conducted in Zone 1, Mangrove Forests of UMT, Terengganu. In this study, two techniques were used: Direct Plating Technique and Damp Incubation Technique. In both techniques the fragments were incubated on Sea Water Agar (SWA) and Potato Dextrose Agar (PDA). More fungi were detected on PDA, indicating that PDA stimulates the growth of fungi. The fungi isolated were subcultured in slide culture technique to identify the fungi. Identification of fungi was based on their morphology and by observation under microscope. A total of 40 individual species of fungi were isolated from both techniques which are 29 from Direct Plating Technique and 11 from Damp Incubation Technique. The fungi isolated include 10 Ascomycetes, 18 Deuteromycetes, one Basidiomycete and 11 unidentified isolates. Out of 29 identified fungi, 28 species belong to terrestrial fungi and one belongs to marine fungi. These fungal isolates can be used further in the investigation of possible bioactive compound(s) produced by these fungi.

**PEMENCILAN DAN PENGECAMAN FUNGI YANG BERASOSIASI
DENGAN *Nypa fruticans* DI UNIVERSITI MALAYSIA
TERENGGANU, TERENGGANU**

ABSTRAK

Tumbuhan paya bakau berpotensi menghasilkan sebatian bioaktif yang berguna untuk tujuan perubatan. Walaubagaimana pun, masih tidak dapat dibuktikan samada sebatian bioaktif itu dihasilkan dari tumbuhan paya itu sendiri atau dari mikrob yang berasosiasi dengan tumbuhan itu. Untuk menentukannya, fungi yang berasosiasi dengan *Nypa fruticans* telah dipencarkan. Persampelan fragmen daun, pelepasan daun dan kulit kayu *N. fruticans* ini dijalankan di Zon 1, Hutan Paya Bakau UMT, Terengganu. Dalam kajian ini, dua teknik yang berbeza telah dijalankan iaitu teknik ‘direct plating’ dan teknik ‘damp incubation’, kedua-duanya menggunakan agar air laut (SWA) dan agar kanji ubi kentang (PDA). Lebih banyak fungi dikesan pada PDA, menunjukkan PDA merangsang pertumbuhan fungi. Fungi yang dipencarkan kemudian disubkulturkan dalam teknik ‘slide culture’ bagi pengecaman fungi yang ditemui. Pengecaman dijalankan berdasarkan morfologi fungi itu dan melalui pemerhatian di bawah mikroskop. Sejumlah 40 individu fungi telah dipencarkan dari kedua-dua teknik dimana 29 fungi dipencarkan dari teknik ‘direct cultur’ dan 11 dari teknik ‘damp incubation’. Fungi yang dipencarkan terdiri daripada 10 Ascomycetes, 18 Deuteromycetes, satu Basidiomycetes dan 11 tidak dapat dicam. Daripada 29 fungi yang dikenalpasti, 28 spesies adalah fungi daratan dan satu adalah fungi marin. Penciran fungi ini boleh digunakan seterusnya dalam kajian penghasilan sebatian bioaktif yang mungkin dihasilkan oleh fungi-fungi ini.