

ANALYSIS OF
INTERNAL ACTIVITY OF
CROWN POLYMERIC CULTURES

GITA SETIA

FAKULTAS SAINS DAN TEKNOLOGI
UNIVERSITAS PANCASILA JAKARTA
2007

© 2012 Pearson Education, Inc.

1100051122

Perpustakaan Sultanah Nur Zahirah (UMT)
Universiti Malaysia Terengganu



LP 9 FST 2 2007



1100051122

Anti vibrio bacteria activity of Cryptocoryne ciliate cultures / Cynthia Seta.

**PERPUSTAKAAN
UNIVERSITI MALAYSIA TERENGGANU (UMT)
21030 KUALA TERENGGANU**

1100051122

Lihat sebelah

HAK MILIK
PERPUSTAKAAN UMT

**ANTI *VIBRIO* BACTERIA ACTIVITY OF
Cryptocoryne ciliata CULTURES**

By

Cynthia Seta

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
UNIVERSITI MALAYSIA TERENGGANU
2007

1100051122

This project should be cited as:

Cynthia, S. 2007. Anti *Vibrio* bacteria activity of *Cryptocoryne ciliata* cultures.
Undergraduate thesis, Bachelor of Science in Biological Sciences, Faculty of
Science and Technology, Universiti Malaysia Terengganu, Terengganu. 56p.

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

UNIVERSITI MALAYSIA TERENGGANU

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

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: ANTI VIBR/O BACTERIA ACTIVITY OF *Cryptocoryne ciliata* CULTURES oleh CYNTHIA SETA, no. matrik: UK10748 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. AZIZ AHMAD
Nama: Pensyarah
Cop Rasmi: Jabatan Sains Biologi
Fakulti Sains dan Teknologi
Universiti Malaysia Terengganu
21030 Kuala Terengganu.

Tarikh: 30/04/2007

Penyelia Kedua / Co-Supervisor

Nama: **DR. NAJIAH MUSA @ ZAKARIA**
Cop Rasmi: Pensyarah
Jabatan Sains Perikanan dan Akuakultur
Fakulti Agroteknologi dan Sains Makanan
Universiti Malaysia Terengganu
21030 Kuala Terengganu

Tarikh: 30/04/2007

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: 30/04/2007

AKNOWLEDGEMENT

First of all thanks be to God for his mercy I am able to complete this project. I particularly grateful to Dr. Aziz Ahmad, my project supervisor, who is devoted his time and effort to guide and advised me throughout my project. I also dedicate my special appreciation to Dr. Najiah Musa, my co-supervisor, who undoubtfully donates the bacteria that being used in this project and her willingness to give guidance and advice throughout my project.

This special thanks also goes to our laboratory assistant, Mr. Mazrul, Mdm. Zarina and Mdm Fatimah for their cooperation and knowledge during my laboratory work. And also not forget to Biological Department of Science Officer Miss Norazlina Abd. Aziz.

I wish to extend my appreciation to the person who has directly or indirectly guiding me throughout the project, especially post graduate students.

Special thanks to my family for their support, encouragement and patience.

Finally, but most importantly, I would like to thank my friends for their cooperation and support to me not only for this project, but also their support and encouragement throughout my studies. I must admit without their help I will unable to complete this report in flying colors.

TABLE OF CONTENTS

| | Page |
|---|-------------|
| ACKNOWLEDGEMENT | ii |
| LIST OF TABLES | v |
| LIST OF FIGURES | vi |
| LIST OF ABBREVIATIONS/SYMBOLS | vii |
| LIST OF APPENDICES | viii |
| ABSTRACT | ix |
| ABSTRAK | x |
| | |
| CHAPTER 1 INTRODUCTION | 1 |
| 1.1 <i>Cryptocoryne ciliata</i> | 1 |
| 1.2 The Important of Study | 1 |
| 1.3 Objectives of the Study | 2 |
| | |
| CHAPTER 2 LITERATURE REVIEW | 3 |
| 2.1 Bacterial Disease | 3 |
| 2.1.1 Bacterial Disease Against Human | 3 |
| 2.1.2 Bacterial Disease Against Aquatic Organism | 4 |
| 2.2 Plant Secondary Metabolites | 4 |
| 2.2.1 Bioactive Properties and use of Araceae Family | 6 |
| 2.3 <i>Vibrio</i> Bacteria | 12 |
| 2.3.1 <i>Vibrio</i> Bacteria Disease | 13 |
| 2.4 Plant Cell Culture: Accumulation of Secondary Metabolites | 14 |
| 2.5 Development of Antibacterial Agents | 15 |
| 2.5.1 Physical Agents of Microbial Control | 15 |
| 2.5.2 Chemical Agents of Microbial Growth | 16 |
| | |
| CHAPTER 3 METHODOLOGY | 17 |
| 3.1 Plant Material | 17 |
| 3.2 Aqueous Extraction | 17 |
| 3.3 Methanol Extraction | 17 |

| | | |
|-----------------------------|--|----|
| 3.4 | Bacteria Strains | 19 |
| 3.5 | Preparation of Assay Medium for Bacteria | 20 |
| 3.6 | Qualitative Antibacterial Bioassay | 20 |
| 3.7 | Quantitative Antibacterial Biossay | 21 |
| 3.8 | Statistical Analysis | 21 |
| CHAPTER 4 RESULTS | | 22 |
| 4.1 | Crude Extraction | 22 |
| 4.2 | Anti <i>Vibrio</i> Bacteria Activity | 23 |
| CHAPTER 5 DISCUSSION | | 25 |
| CHAPTER 6 CONCLUSION | | 29 |
| REFERENCES | | 30 |
| APPENDICES | | 39 |
| VITAE | | 46 |

LIST OF TABLES

| Table | | Page |
|--------------|---|-------------|
| 2.1 | The summary of bioactive properties and other usages of Araceae family plants. | 10 |
| 4.1 | Dry weight and colors of methanol extracts obtained. | 22 |
| 4.2 | Dry weight and colors of aqueous extracts obtained. | 22 |
| 4.3 | Anti <i>Vibrio</i> bacteria activity of <i>C. ciliata</i> methanol extracts (500 μ g/ml). | 23 |
| 4.4 | Anti <i>Vibrio</i> bacteria activity of <i>C. ciliata</i> aqueous extracts (500 μ g/ml). | 23 |

LIST OF FIGURES

| Figure | | Page |
|---------------|---|-------------|
| 3.1 | <i>In vitro</i> culture of <i>Cryptocoryne ciliata</i> . (a) Plant of 30 days, (b) Plants of 50 days (c) Plant of 70 days. | 18 |
| 3.2 | The <i>Vibrio</i> bacteria cultures on the thiosulphate citrate bile salt sucrose (TCBS) (Merck) media. (a) <i>V. vulnificus</i> , (b) <i>V. alginolyticus</i> , (c) <i>V. parahaemolyticus</i> . | 19 |
| 4.1 | Inhibition zones of 30-days, 50-days and 70-days extract against tested (a) <i>V. alginolyticus</i> , (b) <i>V. vulnificus</i> and <i>V. parahaemolyticus</i> . | 24 |

LIST OF ABBREVIATIONS/SYMBOLS

| | |
|---------------------|--|
| BAP | 6-Benzylaminopurine |
| DMSO | Dimethyl sulfoxide |
| MHA | Mueller Hinton agar |
| TSB | Tryptic soy broth |
| TSA | Tryptic soy agar |
| TCBS | Thiosulphate citrate bile salt sucrose |
| L | Liter |
| ml | Milliliter |
| μl | Microliter |
| mm | Millimeter |
| nm | Nanometer |
| g | Gram |
| mg | Milligram |
| μm | Microgram |
| mg/L | Milligram per liter |
| mg/ml | Milligram per milliliter |
| $^{\circ}\text{C}$ | Degree Celsius |
| CFUmL ⁻¹ | Colony forming units per milliliter |
| % | Percentage |

LIST OF APPENDICES

| Appendices | Page |
|---|-------------|
| APPENDIX A DATA ANALYSIS | |
| Table A.1 Inhibition zone diameters (mm) of methanol extract tested disc (500 μ g/ml) for three replicates. | 39 |
| Table A.2 Data analysis of bacteria <i>V. alginolyticus</i> susceptibility to methanol extract by using ANOVA-Oneway. | 40 |
| Table A.3 Data analysis of bacteria <i>V. vulnificus</i> susceptibility to methanol extract by using ANOVA-Oneway. | 43 |

ABSTRACT

Cryptocoryne ciliata is the aquatic plant that easy to propagate *in vitro*, but no report on its medicine value. Thus, the objectives of this study were to determine the anti *Vibrio* bacteria activity and MIC (minimal inhibitory concentration) of extract for antibacterial activity and also to determine the age of plant with highest antibacterial activity. The anti-*Vibrio* bacteria activity of methanol and aqueous extract of *Cryptocoryne ciliata* cultures were investigated. The *in vitro* plantlet was established and proliferate on the MS media added with 3mg/l BAP for 30, 50 and 70 days of cultivation time. The evaluation of antibacterial activity was done by using Kirby-Bauer method. The methanol extract of cultured plants have antibacterial against *V. alginolyticus* and *V. vulnificus* with the diameter in range of 6mm to 9mm, but no antibacterial activity when tested against *V. parahaemolyticus*. The aqueous extracts have no antibacterial activity against three tested bacteria. Among the three cultivation time, 70-days have the highest inhibition zone following by 50 days and 30 days the lowest inhibition zone. These observations indicate of that the longer cultivation time gave higher antibacterial activity. Therefore, the longer cultivation time could increase the yield of plant secondary metabolites that contained antibacterial constituents. Further study should be carried out to obtain the compound responsible for the antibacterial activity.

ANTI-VIBRIO BAKTERIA AKTIVITI BAGI KULTUR *Cryptocoryne ciliata*

ABSTRAK

Cryptocoryne ciliata adalah tumbuhan akuatik yang mudah ditumbuhkan secara *in vitro*, tetapi tiada laporan tentang nilai perubatannya. Oleh itu, objektif kajian ini adalah mengenalpasti anti *Vibrio* bateria aktiviti serta minimum kepekatan perencat dan juga mengenalpasti umur tumbuhan yang mempunyai anti *Vibrio* bacteria yang tinggi. Anti *Vibrio* bakteria aktiviti bagi ekstrak methanol dan air telah dilakukan terhadap tumbuhan kultur *Cryptocoryne ciliata*. Tumbuhan ini ditumbuhkan secara *in vitro* pada media MS ditambah 3mg/l BAP bagi 30, 50 dan 70 hari pengkulturan. Pentaksiran antibakteria telah dijalankan dengan menggunakan kaedah Kirby-Bauer. Esktrak metanol menunjukkan antibakteria aktiviti terhadap *V. alginolyticus* dan *V. vulnificus* dengan julat diameter zon perencat diantara 6mm kepada 9mm, dan tiada antibakteria aktiviti terhadap *V. parahaemolyticus*. Ekstrak air tiada kesan antibakteria aktiviti terhadap kesmua bakteria yang diuji. Ekstrak metanol 70 hari menunjukkan antibakteria yang tinggi berbanding 50 hari diikuti 30 hari pengkulturan. Keputusan ini menunjukkan tempoh pengkulturan yang panjang memberikan kesan antibakteria yang tinggi. Oleh itu, tempoh pengkulturan yang panjang dapat meningkatkan hasil metabolismik sekunder tumbuhan yang mempunyai sebatian antibakteria. Kajian selanjutnya harus dilakukan untuk memperolehi sebatian yang mempunyai antibakteria aktiviti.