UNIVERSITI PUTRA MALAYSIA

ANTIBACTERIAL ACTIVITY OF LOCALLY ISOLATED THERMOPHILIC FUNGI

AKMAL HAYAT ABDUL KARIM

FBSB 2015 91

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Dissertation submitted in partially fulfilment of the

Requirement for the course BMY 4999 Project in the

Department of Microbiology

Universiti Putra Malaysia

June 2015

PENGESAHAN

Dengan ini adalah disahkan bahawa tesis projek yang bertajuk "Antibacterial Activity Of Locally Isolated Thermophilic Fungi" telah disiapkan serta dikemukakan kepada Jabatan Mikrobiologi oleh Akmal Hayat Bin Abdul Karim (162579) sebagai syarat untuk kursus BMY 4999 Projek.

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ACKNOWLEDGEMENTS

In the name of Allah, The Most Gracious, The Most Merciful.

All praises to Allah Almighty, for the strengths and guidance in completing my Final Year Project BMY 4999. Without His help, ones cannot reach their destination.

First and foremost, I would like to express my deepest gratitude to my supervisor, Dr. Wan Zuhainis Saad for her support, guidance and encouragement throughout the completion of this project. I would also express my deepest appreciation to Mohd Khairil Radzali for all the guidance and knowledge in assisting me throughout the year and also to all postgraduate students in Mycology laboratory.

Extend deepest gratitude and devotion to my family for their understanding and support, and also to Coordination Officer PALAPES UPM (Laut), Lt. Kdr. Mohd Syahirudin Bin Hamsin TLDM for the understanding my duty and responsibility as final year student instead of being a part of the Royal Malaysian Navy (RMN) officers.

Lastly, I wish to express my thankful to all those who have been involved in completing this project.

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LIST OF ABBREVIATIONS

°C	-	degree Celcius
µg	-	microgram
g	-	gram
L	-	Litre
mg	-	milligram
mL	-	millimeter
sp	-	species
SSF	-	Solid State Fermetation
SmF	-	Submerged Fermentation
PDA	-	Potato Dextrose Agar
PDB	-	Potato Dextrose Broth
MHA	-	Muller-Hinton Agar
TSB	-	Trypticase Soy Broth

ABSTRACT

Sampling of thermophilic fungi was done at Cherana Puteh Hot Springs in Alor Gajah, Melaka. Isolation was done and two pure cultures were obtained named CP1 and CP2. Then, it was incubated at three different ranges of temperature to determine the best temperature of fungal growth. After that, the growth profiles of isolated thermophilic fungi was measured based on their dry cell weight. The stationary phase growths of thermophilic fungi were determined. For the screening of antibacterial properties, the secondary metabolites of thermophilic fungal secondary metabolites were extracted using methanol. The methanolic mixtures were filtered to separate the filtrate and mycelium. The crude extracts were screened for antibacterial activity against some clinical strains of test microorganisms. The test microorganisms were *Escherichia coli* ATCC 8739, *Escherichia coli* H7 ATCC E187, *Staphylococcus aureus* ATCC 6538, *Pseudomonas aeruginosa* ATCC 15442, *Bacillus subtilis* ATCC 6633, *Salmonella typhimurium* S836, Methicillin-resistant *Staphylococcus aureus* S547, and *Listeria monocytogenes* L10. Disc diffusion method was conducted to screen for the antibacterial properties that produced by the isolates against stated test microorganisms.

ABSTRAK

Sampel *thermophilic fungi* telah berjaya diambil dari Kolam Air Panas Cherana Puteh di Alor Gajah, Melaka. Pengasingan sampel telah selesai dilakukan dan mendapat dua kultur tulen yang dinamakan sebagai CP1 dan CP2. Kemudian ia telah dikulturkan di tiga suhu yang berbeza untuk mengenal pasti suhu terbaik untuk pembesarannya. Selepas itu, pembesaran kultur *thermophilic fungi* diukur di melalui peningkatan berat kering sel dengan. Fasa tumbesaran pegun *thermophilic fungi* dikenal pasti. Untuk saringan keupayaan antimikrob, metabolit sekunder *thermophilic fungi* diekstrak dengan menggunakan metanol dan kandungannya telah ditapis bagi mengasingkan miselium. Ekstrak mentah telah disaring keupayaan antimikrobnya melalui ujian terhadap mikroorganisma sasaran. Kesemua mikroorganisma sasaran adalah *Escherichia coli* ATCC 8739, *Escherichia coli* H7 E187, *Staphylococcus aureus* ATCC 6538, *Pseudomonas aeruginosa* ATCC 15442, *Bacillus subtilis* ATCC 6633, *Salmonella typhimurium* S836, Methicillin-resistant *Staphylococcus aureus* S547, and *Listeria monocytogenes* L10. Kaedah resapan cakera telah diaplikasi untuk mengenal pasti keupayaan antimikrob yang dihasilkan oleh *thermophilic fungi.*

CHAPTER 1

INTRODUCTION

1.1 Research Background

According to Cooney & Emerson (1964), thermophilic fungi are the fungi that have the ability to grow at temperatures between 20°C and 50°C. The distribution of thermophilic fungi occurs at warmer sites. Thermophilic fungi are often distribute in large scales in natural geothermal sites such as hot springs, geothermal soils and hot springs effluent channels (Ellis, 1980).

Thermophilic fungi are special microorganisms with their extremely stable cellular component together with unique metabolic capabilities. This is a great interest to microbiologists and biotechnologists, as the organisms isolated from these environments are a good source of thermostable enzymes and antimicrobial properties offer considerable promise for biotechnological application (Sharma *et al*., 2013).

Thermophilic fungi are productive and talented producers of structurally diverse bioactive metabolites and have yielded some of the most important products for the pharmaceutical industry (Ola, 2014). According to Bérdy (2005), eukaryotic fungi are the most frequent and most versatile producers for bioactive metabolites. Fungal secondary metabolites have revolutionized the yielding of antimicrobial drugs (Aly *et al*., 2011).

Antibiotics are the most important category of secondary metabolites extracted from microorganisms using fermentation (Subramaniyam and Vimala, 2012). The first antibiotics, penicillin G from *Penicillium notatum* was discovered by

Alexander Fleming in 1928 (Aly *et al*, 2011). Antibiotics are products of fermentation and are produced in the stationary phase, often in response to stress conditions (Robinson *et al*., 2001).

The 21st Century opened a new era of antibiotic research. The value of biopharmaceuticals was estimated to be 41 billion dollars in the global market, with a growth rate of 21% over the period from 2004 to 2008. This is due to urgent clinical needs and the increasing serious health problem such as HIV, multi-resistant strains, reappearing mycobacteria thus required development of new technologies which are more effective (Aly *et al*, 2011).

1.2 Problem Statement

Serious infections caused by bacteria that have become resistant to commonly used antibiotics have become a major global healthcare problem in the 21st century. They are not only more severe, require longer and complex treatments, but they are also significantly more expensive to diagnose and to treat (Alanis, 2005).

To date, certain antibacterial agent or antibiotics available in market are irrelevant to be used. The morbidity and mortality has increased including the cost of healthcare due to emergence of antibiotics resistance microorganisms.

Antibiotic resistance, initially a problem of the hospital setting associated with an increased number of hospital acquired infections usually in critically ill and immunosuppressed patients, has now extended out to the community causing severe infections difficult to diagnose and treat (Alanis, 2005).

1.3 Hypothesis.

1. Thermophilic fungi isolated locally from the hot springs may contain antibacterial properties.

1.4 Objectives

1. To isolate local thermophilic fungi
2. To screen the antibacterial properties of thermophilic fungi.

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