UNIVERSITI PUTRA MALAYSIA

ESTABLISHING STANDARD WORKING PROCEDURES TO INVESTIGATE EFFECTS OF CHANNA STRIATUS BLOCH CRUDE EXTRACT ON MALASSEZIA FURFUR GROWTH

FATEMEH EHTESHAM FAR

FPSK(m) 2012 39

ESTABLISHING STANDARD WORKING PROCEDURES TO INVESTIGATE EFFECTS OF *CHANNA STRIATUS* BLOCH CRUDE EXTRACT ON *MALASSEZIA FURFUR* GROWTH

By

FATEMEH EHTESHAM FAR

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirements for the Degree of Master of Sciences.

August 2012

DEDICATION

This thesis is dedicated to all of the following persons who have accompanied me in this way:

My husband, Sadegh A., without Whose Caring Support it would not have been possible

,and

To my Grandmother & Grandfather who support and encouraged me

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Master of Science

ESTABLISHING STANDARD WORKING PROCEDURES TO INVESTIGATE EFFECTS OF *CHANNA STRIATUS* BLOCH CRUDE EXTRACT ON *MALASSEZIA FURFUR* GROWTH

By

FATEMEH EHTESHAM FAR

August 2012

Chairman: Professor Abdul Manan Mat Jais, PhD

Faculty: Medicine and Health Sciences

Skin fungal infections are a common type of skin diseases that has become a global problem. Fungal infections commonly constitute a health problem in developing countries particularly in tropical and subtropical regions. *Malassezia* and *Candida* species are the most prevalent pathogens in human and animals. Yeast-like fungus *Malassezia furfur* is associated with several dermatological diseases such as *Pityriasis versicolor*, *Malassezia* folliculitis, Atopic dermatitis and Seborrheic dermatitis (Dandruff). The available drugs known as antimycotic act against extensive range of molecular targets of fungi but the numbers of antifungal resistance pathogens have increased. Furthermore, majority of these antifungal drugs are

synthetic that lead to some health-threatening side-effects. Consequently, increasing demand for seeking the natural therapeutic medicines has made much attention to pelagic organisms, especially, fish that can be found throughout the ocean. There are no standard parameters for minimum fungicidal concentration (MFC), as well as definitive guideline for evaluating the antifungal drugs against *Malassezia* species. So, this study attempted to apply some modified methods which are more adjustable for *M. furfur* in order to perform antifungal susceptibility tests. Therefore, this research was planned to evaluate effects of *Channa striatus* crude extract on yeast-like fungi *M. furfur* ATCC 14521 through bioassays. Bioassay studies demonstrated that *Channa striatus* crude extract in various concentrations did not show inhibitory activity against *M. furfur* which was evidenced in terms of minimum inhibitory concentration (MIC), minimum fungicidal concentration (MFC) tests and percentage of cell viability using colorimetric assay. Besides, colorimetric resazurin microtitre assay (REMA) Plate Method expressed *Channa striatus* crude extract to have proliferative effect on *M. furfur* in comparison with growth control. As a conclusion, it was previously hypothesized that possibly *Channa striatus* crude extract has sufficient antifungal properties to inhibit M. *furfur* growth. But, based on the *in vitro* data of this study, *Channa striatus* crude extract exacerbate the diseases associated with M. *furfur*. Therefore, it cannot be an effective natural antimycotic medicine to treat skin diseases related to M. *furfur*.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

PEMBANGUNAN STANDARD PROSEDUR KERJA DALAM MENYIASAT KESAN EKSTRAK MENTAH *CHANNA STRIATUS* PADA PERTUMBUHAN *MALASSEZIA FURFUR*

Oleh

FATEMEH EHTESHAM FAR

Ogos 2012

Pengerusi: Profesor Abdul Manan Mat Jais, PhD

Fakulti: Perubatan dan Sains Kesihatan

Jangkitan kulit oleh kulat amat mudah terjadi dan menjadi satu masalah global terutamanya di negara-negara membangun di kawasan-kawasan tropika dan subtropika. Spesies *Malassezia* dan *Candida* adalah agen jangkitan yang paling lazim ke atas manusia dan haiwan. *Malassezia furfur* adalah sejenis yis yang dikaitkan dengan beberapa penyakit dermatologi seperti *versicolor Pityriasis*, *Malassezia* folliculitis, dermatitis atopik dan dermatitis Seborrheic (Kelemumur). Ubatan yang sedia ada dikenali sebagai ubat 'antimycotic' bertindak terhadap pelbagai sasaran molekul di dalam kulat tetapi bilangan patogen yang resistan kepada dadah tersebut telah meningkat. Tambahan pula, kebanyakan ubat antikulat sintetik membawa kepada beberapa kesan sampingan yang boleh mengancam kesihatan. Oleh yang demikian, permintaan yang semakin meningkat untuk menemui ubat-ubatan semula

jadi terapeutik telah membawa perhatian dan kajian ke atas organisma pelagik terutamanya ikan yang boleh didapati di seluruh lautan. Permasalahan timbul apabila tiada garis panduan yang jelas bagi menentukan Kepekatan Fungisidal Minima (MFC) ke atas spesies *Malassezia* agar keberkesanan ubat-ubatan semula jadi terapeutik ini dapat dinilai secara muktamad. Jadi, kajian ini cuba untuk menggunakan beberapa kaedah yang diubahsuai untuk dijalankan ke atas spesies *Malassezia*. Oleh itu, penyelidikan ini telah dirancang untuk menilai kesan ekstrak mentah *Channa striatus* ke atas yis penyebab kelumumur iaitu *M. furfur* ATCC 14521 untuk menilai sifat 'antimycotic' ekstrak mentah *Channa striatus* melalui beberapa kaedah bioassei. Kajian yang dijalankan menunjukkan bahawa ekstrak mentah *Channa striatus* pada kepekatan berlainan tidak menunjukkan aktiviti prencat kulat terhadap *M. furfur* yang telah dibuktikan dari segi kaedah Kepekatan perencatan Minima (MIC), Kepekatan Fungisidal Minima (MFC) dan peratusan sel viabiliti menggunakan kolorimetrik asai. Selain itu, kaedah kalorimetrik resazurin mikrotitre asai (REMA) menunjukkan ekstrak mentah *Channa striatus* mempunyai kesan proliferatif ke atas *M. furfur* berbanding dengan experimen kawalan. Sebagai kesimpulan, hipotesis awal adalah berkemungkinan ekstrak mentah *Channa striatus* mempunyai ciri-ciri anti-kulat yang mencukupi untuk menghalang pertumbuhan *M. furfur*. Tetapi, berdasarkan data *in vitro* dalam kajian ini, didapati ekstrak mentah *Channa striatus* berpotensi untuk menggalakkan lagi pertumbuhan *M. furfur* di dalam sesuatu jangkitan kulit yang berkaitan. Jadi, ia tidak boleh menjadi ubat semulajadi yang berkesan untuk merawat penyakit kulit yang berkaitan dengan *M. furfur*.

ACKNOWLEDGEMENTS

First and foremost I offer my sincerest to the chairman of my supervisory committee; Professor Dr. Abdul Manan B. Mat Jais, who has supported me throughout my research project with his patience and knowledge whilst allowing me to complete this work. I attribute the level of my master degree to his encouragement and effort and without him this thesis, too, would not have been done. One simply could not wish for a better or friendlier supervisor.

I am also very grateful to my co-supervisor; Dr. Mohd Nasir Mohd Desa, who with his kindness, affection, and support gave me courage and ability to overcome all the problems I have faced during course of my study.

Special thanks to the Ministry of Science of Malaysia and Faculty of Medicine and Health Sciences for providing the financial means and laboratory facilities.

Special appreciations to the Institute of Bioscience (IBS) of UPM for providing the laboratory facilities.

The author would also like to convey thanks to great Malaysia; for its hospitality and smiles of its people.

Lastly, I also want to take this opportunity to dedicate my appreciation to my beloved husband, Grandmother, Grandfather and friends for their understanding and endless love, throughout the duration of my study.

I certify that a Thesis Examination Committee has met on 27 August 2012 to conduct the final examination of Fatemeh Ehtesham Far on her thesis entitled " Establishing Standard Working Procedures to Investigate Effects of *Channa striatus* BLOCH Crude Extract on *Malassezia furfur* Growth" in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Master of Science.

Members of the Thesis Examination Committee were as follows:

Sabrina binti Sukardi, PhD
Associate Professor (Medical)
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Chairman)

Malina binti Osman, PhD
Associate Professor
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Internal Examiner)

Roslida binti Hamid @ Abdul Razak, PhD
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Internal Examiner)

Ridzwan Hashim, PhD
Professor
Kulliyyah of Scince
International Islamic University Malaysia
(External Examiner)

SEOW HENG FONG, PhD
Professor and Deputy Dean
School of Graduate Studies
Universiti Putra Malaysia

Date:

This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfillment of the requirement for the degree of Master Science. The members of the Supervisory Committee were as follows:

Abdul Manan Mat Jais, PhD
Professor
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Chairman)

Mohd Nasir Mohd Desa, PhD
Senior Lecturer
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Member)

_____

BUJANG BIN KIM HUAT, PhD
Professor and Dean
School of Graduate Studies
Universiti Putra Malaysia

Date:

DECLARATION

I declare that the thesis is my original work expect for quotations and citations which have been duly acknowledged. I also, declare that it is has not been previously, and is not concurrently, submitted for any other degree at Universiti Putra Malaysia or at other institution.

FATEMEH EHTESHAM FAR

Date: 27 August 2012

TABLE OF CONTENTS