

**ISOLATION AND CHARACTERIZATION OF *ACTINOBACTERIA* FROM
SOIL SAMPLES OF SIGNY ISLAND, ANTARCTICA**

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**DISSERTATION SUBMITTED IN FULFILMENT OF THE REQUIREMENTS
FOR THE DEGREE OF MASTER OF BIOTECHNOLOGY**

INSTITUTE OF BIOLOGICAL SCIENCES

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UNIVERSITY OF MALAYA

KUALA LUMPUR

MALAYSIA

2010

ABSTRACT

In this study, ninety five actinobacterial isolates were cultured from seven soils samples collected on Signy Island, maritime Antarctica. Nine different isolation media were used and the isolates plates were incubated at 15 °C for up to four months. SCN agar serves as the better media for the isolation of actinobacteria. Presence of actinobacteria in the soil samples was also detected using PCR with actinobacteria specific primers. The actinobacterial isolates were characterized based on morphology (Gram-stain, coverslip examination, colour grouping), analysis of DAP isomers, antibacterial activities and by molecular methods (ARDRA, analysis of 16S rRNA sequence and detection of NRPS gene).

The isolates were dereplicated using the colour grouping method which resulted in fifteen colour groups ranging from brownish gray, orange, yellow to white. Representatives from the colour groups were selected for DAP analysis: twenty nine isolates contained LL-DAP while seventeen had *meso*-DAP in their whole cell wall hydrolysates. All ninety five actinobacterial isolates were also dereplicated into sixteen ARDRA groups. Phylogenetic analysis of the 16S rRNA gene sequences from representatives of ARDRA group showed close relationships with the members of the genera *Demetria*, *Glaciibacter*, *Humicoccus*, *Kocuria*, *Marmoricola*, *Mycobacterium*, *Nocardia*, *Rhodococcus*, *Streptomyces* and *Tsukamurella*.

Antimicrobial activity was assayed against six tester bacteria. Forty six actinobacterial isolates inhibited the growth of *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Proteus vulgaris*. NRPS genes were detected in seventy nine isolates and most of these isolates showed antibacterial activity. This study shows the presence of novel actinobacteria in maritime Antarctic soils and Antarctic actinobacteria may provide a new source for antibacterial compounds.

ABSTRAK

Dalam kajian ini, sembilan puluh lima aktinobakteria strain telah diasingkan daripada tujuh sampel tanah di Pulau Signy, maritim Antartika. Sembilan isolasi media yang berlainan telah digunakan dan piring petri dieramkan pada 15 °C sehingga empat bulan. SCN agar merupakan media yang lebih baik untuk pengasingan aktinobakteria. Aktinobakteria telah didapati dalam sampel tanah dan sembilan puluh lima strain telah dikesan dengan menggunakan PCR dengan aktinobakteria primer yang spesifik. Aktinobakteria telah digolongkan berdasarkan morfologi (pewarnaan Gram, ujian sisip kaca, pengelasan mengikut warna), analisis DAP isomer, aktiviti antimikrobial dan cara molekular (ARDRA, analisis 16S rRNA dan pengesanan NRPS).

Semua strain telah direplikasi berdasarkan kombinasi warna koloni dan dikelaskan kepada lima belas kelas yang memaparkan perang kelabuan, jingga, kuning, putih. Wakil dari setiap pengkelasan warna telah dipilih untuk analisis DAP, dua puluh sembilan strain mengandungi LL-DAP manakala tujuh belas strain mengandungi *meso*-DAP. Semua aktinobakteria telah dikelaskan kepada enam belas ARDRA kelas. Phylogenetik analisis 16S rRNA gene daripada wakil ARDRA menunjukkan persamaan yang tinggi dengan genera *Demetria*, *Glaciibacter*, *Humicoccus*, *Kocuria*, *Marmoricola*, *Mycobacterium*, *Nocardia*, *Rhodococcus*, *Streptomyces* and *Tsukamurella*.

Aktiviti antimikrobial telah dijalankan terhadap enam bakteria ujian. Empat puluh enam isolasi menunjukkan aktiviti terhadap *Staphylococcus aureus*, *Staphylococcus epididymis* dan *Proteus vulgaris*. NRPS gene telah dikesan di tujuh puluh sembilan isolasi dan kebanyakan isolasi ini menunjukkan aktiviti antibakterial. Kajian ini menunjukkan kehadiran aktinobakteria novel di kawasan maritim Antartik dan actinobacteria di Antartika menunjuk potensi dalam mengeluarkan sebatian antimikrobial yang baru

ACKNOWLEDGEMENTS

I would like to show my deepest appreciation to everyone who helped me during this period of my research from June 2007 to October 2008. Without them, this research would not have been a reality.

First and foremost, I would like to show my deepest appreciation to both of my supervisors, Dr. Geok Yuan Annie Tan and Prof. Dr. Irene Tan Kit Ping for their continuous guidance and assistance throughout the duration of this report. The duration of working and learning was tough and interesting and I really enjoyed it.

Special thanks to my friends Yew Wen Chyin, Loh Siu Ning, Tan Pei Chuen, Tan Hui Khim, Toh Sui Peng, Aaron Teo, Lim Cheau Theng and Lim Mei Sim who gave me support and assisted me by providing useful reference material and information throughout this research. I would also like to express my foremost appreciation and gratitude to Teh Wei Ping, who have spent time helping me in my research and been with me through the long laboratory hours.

I would also like to thank Mr. Hafiz and Miss Anieliza in helping to provide laboratory equipments for my research. I would like to give acknowledgement to my pastor's family for their continuous support and prayer. Last but not least, I would like to thank and express my appreciation to my parents and sister for their encouragement, guidance and support.

Parts of this thesis have been presented in conferences:

Pan Shing-Yi, Irene Kit-Ping Tan, Geok-Yuan Annie Tan. (2008). Isolation and characterization of Actinobacteria in soil samples obtained from maritime Antarctica. 30th symposium of Malaysian society of Microbiology (16th – 19th August 2008), Universiti Putra Malaysia, Selangor, Malaysia.

Pan Shing-Yi, Irene Kit-Ping Tan, Geok-Yuan Annie Tan. (2009). Characterization of Culturable Actinobacteria from Maritime Antarctic Soils. 4th Malaysian International Seminar on Antarctic (MISA) (1st – 3rd April 2009), University of Malaya, Kuala Lumpur, Malaysia.

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ABBREVIATIONS

%	:	Percentage
°C	:	Degree Celsius
cm	:	centimetre
<i>et al.</i>	:	exempli gratis (example)
ed.	:	Editor
g	:	Gram
L	:	Litre
mg	:	Milligram
min	:	Minutes
ml	:	Millimetre
No.	:	Number
p.s.i	:	Pound square inch
r.p.m	:	revolution per minute
sp.	:	Species
v/v	:	Volume per volume
w/v	:	Weight per volume
µl	:	Microlitre