MOLECULAR CHARACTERIZATION OF BACTERIA FROM SPONGE, *Theonella* sp.

MOHD NUR FIRDAUS BIN ABDUL LATIF

FAKULTI SAINS DAN TEKNOLOGI
UNIVERSITI MALAYSIA TERENGGANU

2008

Molecular characterization of bacteria from sponge, Theonella sp. / Mohd Nur Firdaus Abdul Latif.

MOLECULAR CHARACTERIZATION OF BACTERIA FROM SPONGE, *Theonella* sp.

By
Mohd Nur Firdaus Bin Abdul Latif

A thesis submitted in partial fulfillment of the requirements for the award of the degree of Bachelor of Science (Biological Sciences)

DEPARTMENT OF BIOLOGICAL SCIENCES
FACULTY OF SCIENCE AND TECHNOLOGY
UNIVERSITY MALAYSIA TERENGGANU
2008

This project should be cited as:

Mohd Nur Firdaus, A.L. 2008. Molecular characterization of bacteria from sponge, *Theonella* sp. Undergraduate thesis, Bachelor of Science in Biological Sciences, Faculty of Science and Technology, Universiti Malaysia Terengganu. 66p.

JABATAN SAINS BIOLOGI
FAKULTI SAINS DAN TEKNOLOGI
UNIVERSITI MALAYSIA TERENGGANU

PENGAKUAN DAN PENGESAHAN LAPORAN PITA I DAN II

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: **Molecular Characterization of Bacteria from Sponge, *Theonella* sp.** oleh **Mohd Nur Firdaus Bin Abdul Latif**, No. matrik: **UK11754** 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:

.....
Penyelia Utama

Nama : DR. NORAZNAWATI BINTI ISMAIL

Cop Rasmi : DR. NORAZNAWATI BINTI ISMAIL
Pensyarah
Jabatan Sains Biologi
Fakulti Sains dan Teknologi
Universiti Malaysia Terengganu
21030 Kuala Terengganu

Tarikh: 21/5/08

.....
Ketua Jabatan Sains Biologi

Nama : PROF. MADYA DR. AZIZ BIN AHMAD

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

Tarikh: 15 JUN 2008

DECLARATION

I, Mohd Nur Firdaus Bin Abdul Latif, hereby declare that this thesis entitled Molecular Characterization of Bacteria from Sponge, *Theonella* sp. is the result of my own research except as cited in the references.

Signature : .....

Name : MOHD NUR FIRDAUS ABDUL LATIF

Matric No. : UKM1754

Date : 10 MEI 2008

ACKNOWLEGDEMENTS

Alhamdulillah, in the name of Allah and with His Blessing, I have finished my final year project and thesis writing. I would like to take this opportunity to give my special thanks to my final year project supervisor, Dr. Noraznawati Ismail who gave me a valuable knowledge in molecular biology research and full commitment in guiding me in my project. Thanks to her ideas, strong support and editorial guidance during the discussion time despite of her very busy time. I would also like to extend my thanks to Dr. Cha Thye San for his invaluable advises and help in term of materials and equipments during my research work.

My sincere thank goes to all the research assistants and master students, Brother Lokman, Zairol, Azmi, Willy, Tevan, Miss Kim, Francy, Anne, Kak Nor and Kak Fizah for their guidance and help during my research work in laboratory especially during the absent of my supervisor. Thank you so much for their great support and demonstration in my project. Many thanks also goes to Cik Azlina and all the laboratory assistants, Mr. Mazrul, Puan Izzy, Puan Ku, Kak Ti and Puan Fatimah for their full cooperation on laboratory management. I also will not forget to express my great feeling working around the other members in the group of Dr. Azna which are Azan, Azie, Nani, Ida, Mai, Sena, Kak Zura, Aini, Maizura, Hanin, Huda, Yan, and Khairul Naim. They have made my working environment more enjoyable and meaningful.

I am grateful to have time working with the other final year students of Dr. Cha, Wilonita, Pei Ling, Soo Yin, Tisya, Jihah and Maliza in Biomolecule laboratory that have sharpen my knowledge in my field of study in term of discussion and exchange the information. I would also like to thank my friends especially my housemates, Zul, Afiq, Hasri, Adli, Raslah, Jas and Faizal who made my life more colourful as university student.

Last but not least, my special thanks goes to my loving family, my father and mother, Abdul Latif Ismail and Sobriah Abdul Aziz, my eldest brother and sister, Nur Hisyam and Fatimatuzahrah, her husband, Zulkifli, and two younger sisters, Nur Atikah and Nur Amirah for their du'a, understanding and tolerance. Thank you.

ABSTRACT

Bacteria already known to have association with the sponges but their roles within the sponges still being unclear. Perhaps, the presence of uncultured bacteria within those sponges have made study by culture-dependent techniques are not suitable. Thus, this present study was focused mainly on identification of bacteria associated with the sponges using molecular approaches and investigation of total community of bacteria within those sponges obtained at Karah Island, Terengganu. Amplification of extracted DNA using RAPD-PCR and analysis of the 16S rDNA clones have been carried out to determine the type of bacteria strains in the sponge samples. As a result, RAPD profile generated the unique polymorphism produced by nine samples of sponge and a total of twenty colonies were obtained in the construction of 16S rDNA clones from sample F due to the low number of transformants during the shorter time of ligation reaction. Analysis of 16S rDNA clones retrieved from sample F that represented the sponge, *Theonella* sp. indicated that 100 % of uncultured bacteria. Three out of twenty colonies which were F2, F6 and F12 have been successfully extracted of its plasmid and sequenced. Result indicated that uncultured bacteria clone TK35 and uncultured cyanobacterium clone 106-2-1 with 94-98 % sequence similarity analyzed after BLAST. Results are in agreement with previous study that showed the abundances of the bacteria community including the uncultured bacteria within the sponges.

ABSTRAK

Bakteria telah diketahui mempunyai perhubungan dengan span namun peranannya di dalam span masih lagi tidak jelas disebabkan keraguan hubungan span-bakteria ini. Barangkali kehadiran bakteria yang tidak boleh dikulturkan dalam span tersebut telah membuatkan kajian menggunakan teknik berdasarkan kultur tidak lagi sesuai. Oleh yang demikian, kajian ini telah memberi fokus terutamanya tentang pengenalpastian bakteria yang berhubungan dengan span tersebut menggunakan pendekatan molekul dan menyelidik jumlah komuniti bakteria di dalam span yang diperolehi dari Pulau Karah, Terengganu. Ekstrak DNA diamplifikasi melalui RAPD-PCR dan analisis klon 16S rDNA telah dijalankan untuk menentukan jenis bakteria yang hadir dalam sampel span. Keputusannya, profil RAPD menunjukkan polimorfisme yang unik dihasilkan oleh sembilan span sampel dan hanya sejumlah dua puluh koloni diperolehi dalam pembinaan klon 16S rDNA dari sampel F berpunca daripada jumlah penukaran sel perumah yang rendah disebabkan masa yang singkat dalam tindak balas penyambungan antara cebisan DNA dengan vektor. Analisis klon 16S rDNA yang diperolehi daripada sampel F yang mewakili span, *Theonella* sp. menunjukkan kehadiran 100 peratus bakteria yang tidak boleh dikulturkan. Tiga daripada dua puluh koloni iaitu berlabel F2, F6 dan F12 telah berjaya diekstrak plasmidnya dan ditentukan jujukan DNA. Keputusan menunjukkan bakteria yang tidak boleh dikultur, klon TK35 dan sianobakteria yang tidak boleh dikultur, klon 106-2-1 dengan persamaan jujukan antara 94 hingga 97 peratus dianalisis selepas BLAST. Keputusan ini menyokong kajian yang dilakukan sebelum ini yang menunjukkan kelimpahan bakteria komuniti dalam span termasuk bakteria yang tidak boleh dikulturkan.