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CHARACTERIZATION AND ANTIBACTERIAL PROPERTIES OF SOIL *Streptomyces*

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By

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NOR ASMARA BINTI TASRIP

October 2012

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This study was to examine distribution of aerial mass colors and antibacterial activities of 39 *Streptomyces* isolates from soil, and potential genetic relation of the phenotypes by Random Amplification of Polymorphic DNA (RAPD) analysis. *Streptomyces griseus* ATCC 10137 strain was also included as a reference strain (n = 40). Identification was done by conventional and molecular approaches (*16S rRNA* and *rpoB* genes sequence homology). These sequences were further analyzed by using UPGMA (Unweighted Pair Group Method with Arithmetic Mean) method to build phylogenetic tree for *16S rRNA* gene. The phylogenetic tree showed a diverse evolutionary pedigree of the isolates in 6 major clusters. Based on International *Streptomycetes* Project 2 (ISP-2) media, the aerial mass colors observed were categorized as Yellow (n = 15), Grey (n = 9), Brown (n = 7), White (n = 6) and Others (n = 3). Antibacterial activities were assessed on Muller-Hinton agar (MHA) and Trypticase Soy agar (TSA) by perpendicular streak method against *Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, *Salmonella* sp.

and *Enterococcus* sp. MHA demonstrated 15 isolates with broad spectrum antibacterial activities and 18 as non-broad spectrum. On the contrary, TSA gave lower proportions with 15 and 9 isolates respectively. A high proportion of isolates with non-White color showed antibacterial activities to suggest a potential correlation. RAPD dendogram (a composite of three random primers) also clustered majority of them but segregated those of White color, which showed less antibacterial activities, in a different cluster. Thus, a correlation between aerial mass color and antibacterial property, with a potential genetic linkage, could be assumed.

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

PENCIRIAN DAN SIFAT ANTIBAKTERIA *Streptomyces* DARI TANAH

Oleh

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Kajian ini adalah untuk mengkaji tentang taburan warna 'aerial mass' dan aktiviti antibakteria oleh 39 *Streptomyces* isolat yang diasingkan daripada tanah, dan potensi hubugan antara maklumat genetik dengan phenotypes oleh analisis menggunakan Amplifikasi DNA Polimorf rawak (RAPD). *Streptomyces griseus* ATCC 10137 strain juga telah dimasukkan sebagai strain rujukan (n = 40). Identifikasi telah dilakukan dengan pendekatan konvensional dan molekular (gen *16S rRNA* dan *rpoB* jujukan homology). Seterusnya jujukan ini dianalisis dengan menggunakan kaedah UPGMA (Pasangan Unweighted Kumpulan dengan Kaedah Aritmetik Min) untuk membina pokok filogenetik untuk gen *16S rRNA*. Pokok filogenetik itu menunjukkan salasilah isolat daripada pelbagai evolusi dalam 6 kelompok utama. Berdasarkan (Antarabangsa *Streptomycetes* Projek 2) ISP-2 media, warna 'aerial mass' yang diperhatikan telah dikategorikan sebagai Kuning (n = 15), Kelabu (n = 9), Coklat (n = 7), Putih (n = 6) dan lain-lain (n = 3). Aktiviti antibakteria telah dinilai menggunakan MHA (Muller Hinton agar) dan TSA (Trypticase agar soya) dengan kaedah streak yang berserenjang dengan *Staphylococcus aureus* ATCC 25923,

Escherichia coli ATCC 25922, *Salmonella sp.* dan *Enterococcus sp.* MHA menunjukkan 15 isolat dengan antibakteria berspektrum luas dan 18 sebagai berspektrum bukan luas. Sebaliknya, TSA menunjukkan aktiviti antibakteria yang lebih rendah dengan masing-masing 15 dan 9 isolat. Sebahagian besar daripada isolat dengan warna bukan Putih menunjukkan aktiviti antibakteria yang berpotensi untuk mencadangkan korelasi. RAPD dendogram (komposit daripada tiga primers secara rawak) juga mengumpulkan majoriti daripada mereka tetapi diasingkan mereka yang mempunyai warna Putih, yang menunjukkan aktiviti kurang antibakteria, dalam kelompok yang berbeza. Oleh itu, korelasi antara warna 'aerial mass' dan aktiviti antibakteria, dengan rangkaian genetik yang berpotensi, boleh diandaikan.

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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 of Sciences. The members of the Supervisory Committee were as follows:

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DECLARATION

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

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