PREPARATION FOR *de novo* SEQUENCING OF *Ganoderma* sp. AFFECTING OIL PALM IN Sabah, MALAYSIA

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DISSERTATION SUBMITTED IN FULFILLMENT FOR THE DEGREE OF MASTER OF SCIENCE

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2012

ABSTRACT

PREPARATION FOR *de novo* SEQUENCING OF *Ganoderma* sp. AFFECTING OIL PALM IN SABAH, MALAYSIA

Four types of preparation were done prior to *de novo* sequencing of *Ganoderma* sp. causing basal stem rot in oil palm located in Sabah, Malaysia. Firstly, at least 12 µg of DNA with the ratio of OD_{260} to OD_{280} between 1.7 and 2.0 was obtained from the mycelia of *Ganoerma* sp. strain C in order to be sent for *de novo* sequencing. Secondly, the DNA sequencing of PCR amplified region of ITS1, 5.8S rDNA and ITS2 of the four strains of *Ganoderma* sp. was conducted, revealing that strain A was actually identical to strain E. Both strains were much closely related to strain B than to strain C. Regardless, all the strains were considered to be of the same species when they were compared to the more well-known *G. lucidum*. BLAST revealed that the identification of the strains could not be done due to the limited genetic information regarding the species in *Genbank* database. All of the sequences were submitted to the database and given the accession numbers, therefore freely allowing others to analyze the DNA sequence in the database. The workflow of SOAP, AbySS and Velvet was prepared and tested with the paired-end sequences of human 18 chromosome 12 to compare the contigs produced by them. Unfortunately, no clones containing DNA of *Ganoderma* sp. were successfully produced. All the preparations were done in anticipation that the assembly and analysis of the *de novo* sequences of *Ganoderma* sp. would be hastened.

ABSTRAK

Empat jenis penyediaan telah dibuat sebelum dijalankannya penjaluran de novo Ganoderma sp. yang menyebabkan pereputan batang dasar dalam kelapa sawit di Sabah, Malaysia. Persiapan yang pertama ialah pengekstrakan sekurang-kurangnya 12 µg DNA yang mempunyai kadar OD_{260} kepada OD_{280} yang terletak antara 1.7 dan 2.0 daripada miselium strain C Gandoerma sp. Penjaluran DNA di kawasan ITS1, 5.8S rDNA dan ITS2 yang diamplifikasikan dengan PCR bagi keempat-empat strain Ganoderma sp. dijalankan dan didapati bahawa strain A adalah sama dengan strain E. Kedua-dua strain ini didapati berhubungan dekat dengan strain B daripada strain C. Walau bagaimanapun, keempat-empat strain tersebut didapati berasal daripada spesies yang sama setelah dibandingkan dengan G. lucidum yang lebih dikenali. BLAST menunjukkan bahawa identiti spesies strain-strain tidak dapat ditentukan kerana kekurangan maklumat genetik tentang spesies tersebut. Semua jaluran DNA diletakkan dalam pangkalan data GenBank dan diberikan nombor ases supaya dapat dianalisasikan oleh orang ramai. Akhirnya, aliran kerja bagi SOAP, ABySS dan Velvet telah disediakan dan diuji dengan jaluran DNA berakhiran pasangan daripada kromosom 12 manusia 18 untuk membuat perbandingan antara kontig-kontig yang dihasilkan. Malangnya, tiada klon-klon yang mengandungi DNA Ganoderma sp. berjaya dihasilkan. Semua persediaan dibuat dengan harapan bahawa pengaturan and penganalisaan jaluran-jaluran de novo Ganoderma sp. dapat dipercepat.