# ISOLATION OF PALMITOYL-ACP THIOESTERASE GENE FROM *Chlorella* sp.

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FAKULTI SAINS DAN TEKNOLOGI
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# ISOLATION OF PALMITOYL-ACP THIOESTERASE GENE FROM *Chlorella* sp.

By
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**DEPARTMENT OF BIOLOGICAL SCIENCES**
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JABATAN SAINS BIOLOGI
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## PENGAKUAN DAN PENGESAHAN LAPORAN PITA I DAN II

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: **Isolation of Palmitoyl-ACP Thioesterase Gene from *Chlorella* sp.** oleh **See Soo Yin**, No. Matrik: **UK12490** 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, UMT.

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## DECLARATION

I hereby declare that this thesis entitled **Isolation of Palmitoyl-ACP Thioesterase Gene from *Chlorella* sp.** is the result of my own research except as cited in the references.

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## ABSTRACT

Palmitoyl-ACP thioesterase (PTE) involves in the synthesis of palmitate (C16:0) from palmitoyl-ACP (16:0-ACP) in fatty acid biosynthesis pathway. The significant role of PTE in this pathway is the basis for the isolation of the gene from *Chlorella* sp. The total RNA extracted from *Chlorella* sp. was reverse transcribed with KPN-T17 oligo-dT primer by using M-MLV reverse transcriptase. Four heterologous forward primers designed from the conserved regions of the PTE gene were used to amplify the corresponding 3'-end regions of the gene. PCR amplification successfully produced five putative DNA fragments with size ranging from 600 bp to 1300 bp. These five putative fragments were cloned into pGEM-T vector and the plasmids extracted from the positive recombinant clones were sent for DNA sequencing to determine their nucleotide sequences. Analysis of sequencing results by searching the GenBank database using BLAST programme revealed that there were no homology to the target PTE gene. Instead, the sequences show homology to other genes, such as eukaryotic translation elongation factor 1 alpha 1 (XP_001696568), aspartate aminotransferase (AAN76499), hypothetical protein (CAN76227), and SocE (AAF91388).

## ABSTRAK

Enzim palmitoyl-ACP thioesterase (PTE) terlibat dalam penghasilan asid palmitik (C16:0) daripada palmitoyl-ACP (16:0-ACP) di dalam mekanisme biosintesis asid lemak. Peranan penting yang dimainkan oleh PTE dalam mekanisme tersebut merupakan sebab utama pemencilan serpihan gen PTE daripada spesies *Chlorella* dilakukan. RNA keseluruhan daripada spesies *Chlorella* telah ditranskripsi berbalik dengan menggunakan primer KPN-T17 oligo-dT dan M-MLV transkriptase berbalik. Empat pencetus yang direkabentuk berasaskan kepada kawasan terpelihara gen PTE telah digunakan untuk mengamplikasikan kawasan gen pada hujung 3' yang sepadan. Amplikasi PCR telah berjaya menghasilkan lima jalur putatif yang bersaiz antara 600 hingga 1300 bp. Kelima-lima jalur putatif ini telah berjaya diklonkan ke dalam vektor pGEM-T dan plasmid diekstrak daripada klon-klon rekombinan yang positif untuk analisis jujukan DNA bagi menentukan jujukan-jujukan nukleotidanya. Hasil analisis jujukan menggunakan pencarian database GenBank menerusi program BLAST menunjukkan tiada homologi terhadap gen PTE. Sebaliknya, jujukan-jujukan tersebut menunjukan homologi terhadap gen-gen lain, antaranya ialah faktor 1 alfa 1 pemanjangan translasi eukaryotik (XP_001696568), aspartat aminotransferase (AAN76499), protein hipotetik (CAN76227), dan SocE (AAF91388).