ISOLATION AND PARTIAL CHARACTERIZATION OF
BETA-KETOACYL-ACP SYNTHASE I (KAS I)
FROM CLOVE FROM *Chlorella* sp.

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ISOLATION AND PARTIAL CHARACTERIZATION OF BETA KETOACYL-ACP SYNTHASE I (KAS I) cDNA CLONE FROM *Chlorella* sp.

By
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PENGAKUAN DAN PENGESAHAN LAPORAN
PROJEK PENYELIDIKAN I DAN II

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: **ISOLATION AND PARTIAL CHARACTERIZATION OF BETA KETOACYL-ACP SYNTHASE I (KAS I) cDNA CLONE FROM *Chlorella* sp.** oleh **NAJIHAH BINTI MOHAMED @ GHAZANI**, no. matrik: **UK12523** 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.

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DECLARATION

I hereby declare that this thesis entitled **ISOLATION AND PARTIAL CHARACTERIZATION OF BETA KETOACYL-ACP SYNTHASE I (KAS I) cDNA CLONE FROM *Chlorella* sp.** is the result of my own research except as cited in the references.

Signature : 

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Date : 11/5/2008

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ABSTRACT

Beta ketoacyl-ACP synthase I (KAS I) is a fatty acid synthase (FAS) that catalyze the elongation of growing fatty acid chains from butyryl-ACP (C4:0) to palmitoyl-ACP (C16:0). In this study, the partial-length of KAS I cDNA clone was isolated and characterized to understand the regulatory mechanism of fatty acid biosynthesis pathway in *Chlorella* sp. at molecular level. Four heterologous forward primers designed from the conserved regions of the KAS I gene were used in combination with KPN-adaptor reverse primer to amplify the corresponding 3'-end region of the gene by using RT-PCR technique. PCR amplification successfully produced five putative DNA fragments with size ranging between 600 bp to 1000 bp. Cloning and sequencing of the fragments revealed that clone designated as pKASI-Ch5 showed significant (74-75%) nucleotide and amino acid sequence identities to KAS I gene from various plant species such as *Helianthus annus*, *Glycine max* and *Arabidopsis thaliana*. The pKASI-Ch5 clone (1008 bp) consists of 639 bp open reading frame (ORF) encoding a partial KAS I polypeptide of 213 amino acids with a 369 bp long 3'-untranslated region. This sequence had been registered in GenBank under accession number EU590913. The isolation of this KAS I cDNA clone is a significant step towards the genetic manipulation of marine microalgae to enhance the production of saturated fatty acids for biodiesel industry.

PEMENCILAN DAN PENCIRIAN SEPARA KLON cDNA BETA KETOASIL-ACP SINTASE I (KAS I) DARIPADA *Chlorella* sp.

ABSTRAK

Beta ketoasil-ACP sintase I (KAS I) merupakan enzim sintase asid lemak (FAS) yang memangkinkan pemanjangan rantaian asid lemak daripada butil-ACP (C4:0) kepada palmitoil-ACP (C16:0). Untuk memahami mekanisme tindakan pengawalaturan biosintesis asid lemak di dalam *Chlorella* sp. pada peringkat molekul, klon separa cDNA KAS I telah dipencil dan dicirikan di dalam kajian ini. Empat pencetus heterologous ke hadapan yang direka berdasarkan kawasan terabadi pada gen KAS I telah digunakan dengan kombinasi primer berbalik KPN-adaptor untuk amplifikasi bahagian 3'-akhiran yang sepadan dalam gen menggunakan teknik RT-PCR. Amplifikasi PCR telah berjaya menghasilkan lima serpihan DNA dengan saiz jangkaan 600 bp hingga 1000 bp. Hasil pengklonan dan penjujukan serpihan DNA mendapati klon pKASI-Ch5 menunjukkan homologi (74-75%) nukleotida dan asid amino yang tinggi dengan gen KAS I daripada pelbagai spesis tumbuhan seperti *Helianthus annus*, *Glycine max* dan *Arabidopsis thaliana*. Klon pKASI-Ch5 (1008 bp) ini terdiri daripada 639 bp panjang rangka bacaan terbuka (ORF) yang mengekod polipeptida KAS I separa dengan 213 asid amino dan 369 bp panjang bahagian 3'-akhiran tidak mengekod. Jujukan ini telah didaftarkan di GenBank di bawah nombor capaian EU590913. Pemencilan klon cDNA KAS I ini merupakan langkah penting dalam manipulasi genetik mikroalga marin untuk penghasilan rantaian asid lemak tepu yang lebih tinggi untuk kegunaan industri biodiesel.