DEVELOPMENT OF SSR - MICRO SATELLITE MARKER
FOR ETSRS GENE FROM Dioscorea hispida

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FACULTY OF BIOTECHNOLOGY
UNIVERSITI MALAYSIA TERENGGANU

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Development of C319-allele specific marker for EPSPS gene from Eleusine indica. / Wilonita Win.

Lihat sebelah

DEVELOPMENT OF C_{319}-ALLELE SPECIFIC MARKER FOR EPSPS GENE FROM *Eleusine indica*

By
Wilonita A/P Win

A thesis submitted in partial fulfillment of
the requirements for the award of the degree of
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DEPARTMENT OF BIOLOGICAL SCIENCES
FACULTY OF SCIENCE AND TECHNOLOGY
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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: **Development of C_{319}- Allele Specific Marker for EPSPS Gene from *Eleusine indica*** oleh **Wilonita A/P Win**, no.matrik: **UK 12861** 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. CHA THYE SAN
Cop Rasmi: Pensyarah
Jabatan Sains Biologi
Fakulti Sains dan Teknologi
Universiti Malaysia Terengganu
21030 Kuala Terengganu.

Tarikh: 4/5/2008

.....
Penyelia Bersama (jika ada)
Nama: DR. CHUAH TSE SENG
CopRasmi: Pensyarah
Jabatan Agroteknologi
Fakulti Agroteknologi dan Sains Makanan
Universiti Malaysia Terengganu
21030 Kuala Terengganu

Tarikh: 5/5/2008

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

Tarikh: 04 MAY 2008

DECLARATION

I hereby declare that this thesis entitled **Development of C_{319}- Allele Specific Marker for EPSPS Gene from *Eleusine indica*** is the result of my own research except as cited in the references.

Signature : .....

Name : WILONITA A/P WIN

Matrix No : UK 12861

Date : 04.05.08

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ABSTRACT

The herbicides resistant weeds become a major threat in Malaysia's plantation. *Eleusine indica* (L.) Gaertn appeared as resistance to glyphosate in 1997. Hence, the development of C_{319} allele specific marker will help in controlling the weed in the effective ways. Genomic DNA extracted from susceptible and different level of resistances of goosegrass was used in this study. EPSPS allele specific clones were generated by cloning of the PCR fragments. EPSPS-C_{319} marker was optimized and used in screening of different resistant level of goosegrass with the EMC-F/ES2-R primer combination. The PCR optimization showed that the optimum condition was at 35 cycles and annealing temperature of 61 °C. Susceptible biotypes showed the presence of C allele in all ten samples. The resistant of 6-fold and 4-fold also showed the existence of C allele at specific band of 628 bp while no band produced in resistant 8-fold. Only C_{319} allele specific marker was successfully developed in this study thus further research on other allele specific marker could be carried out in the future.

ABSTRAK

Kerintangan terhadap racun rumpai menjadi masalah yang utama dalam bidang tanaman di Malaysia. Spesies rumpai, *Eleusine indica* (L.) Gaertn telah dikenal pasti mengalami kerintangan pada racun gylphosate pada tahun 1997. Kajian ini dijalankan dalam membangunkan alel C_{319} sebagai penanda alel spesifik yang penting dalam pengawalan rumpai ini. Pengekstrakan DNA dilakukan pada tahap kerintangan yang berbeza digunakan dalam kajian ini. Klon alel spesifik EPSPS ditentukan oleh proses pengklonan fragmen PCR. Suhu penanda EPSPS-C_{319} dioptimumkan dan digunakan dalam penyaringan rumpai yang mempunyai tahap kerintangan berbeza menggunakan kombinasi primer EMC-F/ES2-R. Pengoptimuman PCR mendapati 35 kitaran dan pada suhu 61 °C adalah yang terbaik. Spesies yang tidak mempunyai kerintangan terhadap racun rumpai digunakan sebagai kawalan dan menunjukkan penghasilan jalur 628 bp pada kesemua sampel. Kerintangan pada tahap 6 dan 4 juga menghasilkan jalur pada 628 bp. Walaubagaimanapun, tiada jalur dihasilkan pada kerintangan tahap 8. Kajian ini hanya berjaya membangunkan penanda spesifik alel C_{319} seterusnya mengharapkan kajian seterusnya akan membangunkan lebih banyak jenis penanda spesifik alel.