



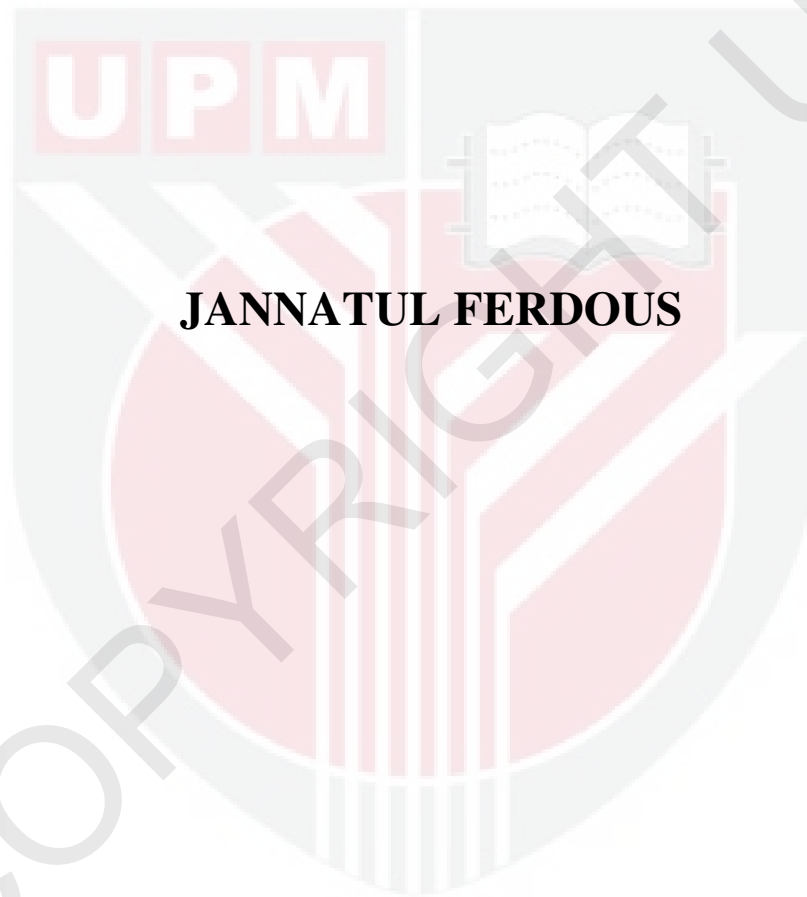
**UNIVERSITI PUTRA MALAYSIA**

***MORPHOLOGICAL, NUTRITIONAL AND MOLECULAR  
CHARACTERIZATION OF SELECTED GENOTYPES FOR UPLAND RICE  
IMPROVEMENT THROUGH MARKER-ASSISTED BACKCROSS BREEDING***

**JANNATUL FERDOUS**

**ITA 2012 5**

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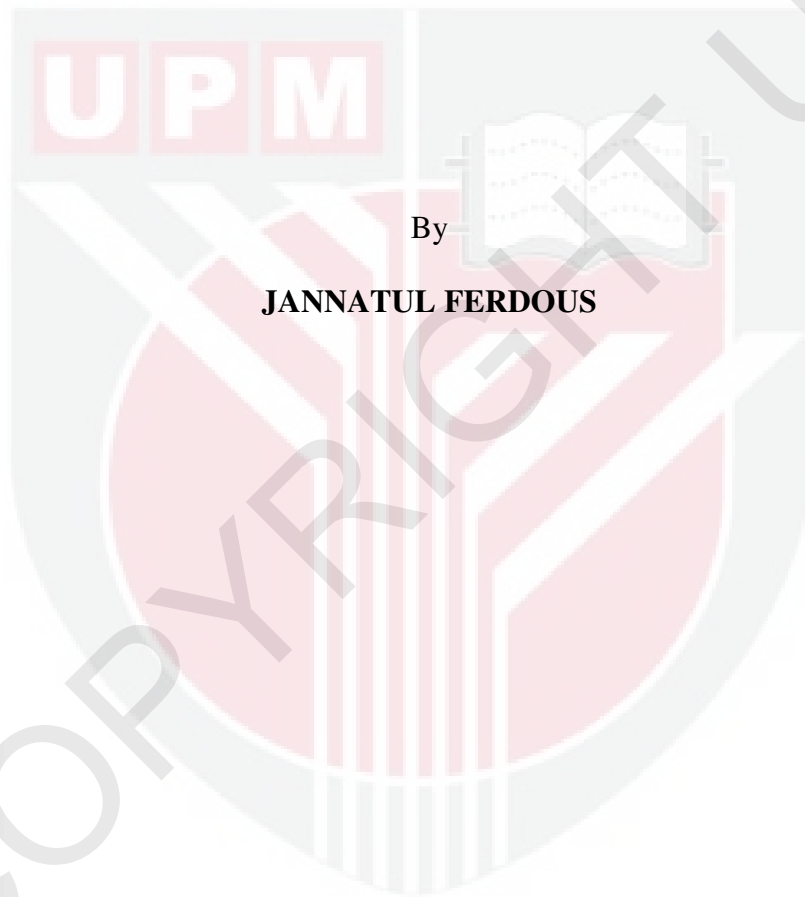


**JANNATUL FERDOUS**

**DOCTOR OF PHILOSOPHY  
UNIVERSITI PUTRA MALAYSIA**

**2012**

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By

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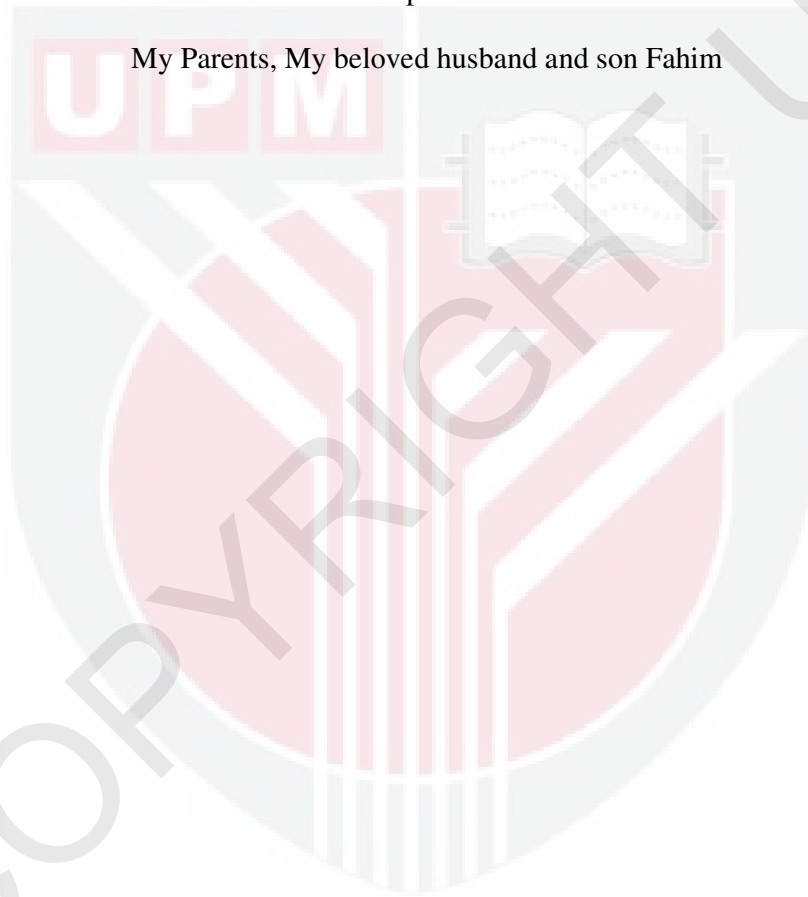


**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia,  
in Fulfillment of the Requirement for the Degree of Doctor of Philosophy**

**April 2012**

## **DEDICATIONS**

This Thesis is Special Dedicated to  
My Parents, My beloved husband and son Fahim



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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Doctor of Philosophy

**MORPHOLOGICAL, NUTRITIONAL, AND MOLECULAR  
CHARACTERIZATION OF SELECTED GENOTYPES FOR UPLAND RICE  
IMPROVEMENT THROUGH MARKER-ASSISTED BACKCROSS  
BREEDING**

By

**JANNATUL FERDOUS**

April 2012

**Chairman : Professor Mohamed Hanafi Musa, PhD, AMIC**

**Institute : Tropical Agriculture**

Upland rice genotypes are low yielder but possess special traits. Marker assisted backcross breeding is a convenient technique for improvement of upland genotype in terms of yield and quality, where desirable trait is selected in early backcross generation. The aims of this study were to introduce high amylose content and high vitamin E alleles in BC<sub>1</sub>F<sub>1</sub> generation as well as select the plant with desirable alleles through marker assisted selection (MAS) using SSR markers. Thirty nine rice genotypes were evaluated for parent selection based on morphology, nutritional composition, and genetic diversity. Three upland genotypes: Bukit Garam582, Bukit Garam1449, and Karingam were selected and back crossed with two high yielding varieties (BR16 and MR219). Identification of desirable plants for amylose content and vitamin E in BC<sub>1</sub>F<sub>1</sub> generation was performed by MAS. The significant variation was observed among the genotypes for yield contributing characters, chlorophyll content, amylose content, glycemic index (GI), and antioxidant activity. The rice genotypes were grouped into 3 clusters, where Pulut Hitam Pahang (PHP), Lansan, and Karibang fell into 3 different solitary positions by morphological clustering. The first four principle components explained 76.5% variation for all

morphological characters and showed positive correlation between tiller number, panicle number, filled grain percentage and grains per panicle. The highest apparent amylose content (27.1%) and the lowest GI (71.1) were observed in BR16. The highest GI (99.2) was found in Bukit Hitam (waxy rice) which contained low amylose (1.5%). The highest antioxidant activity (89.8%) was found in Karingam and the lowest (50.8%) was in BR16. The highest Fe, Zn, and Mg were observed in white and high yielding genotypes. The highest Mn and Cu were detected in local upland black genotype Bukit Hitam (54.4 µg/g) and red genotype Padi Hijau Manis (23.2 µg/g) genotypes, respectively. The genetic diversity was assessed using 25 SSR markers. Cluster analysis showed differentiation of rice genotypes into 4 major groups and several sub-groups. Cross and reciprocal cross in 10 combination between selected five genetically diverse parents were performed. The F<sub>1</sub> plants were confirmed using Wx, RM190, RM3187, and RM3827 markers. The RM190 and Wx markers were used to select waxy allele for high amylose content. The RM3187 and RM3827 were used to select desirable allele of HGGT and HPT, respectively, which are responsible for tocopherols and tocotrienols biosynthesis. The following markers were amplified: for high amylose content (RM190 and Wx at 107 bp fragment); desirable allele of qTOC-6-2 (tocopherol) (RM3187 at 148 bp) and qT3-6-1 (tocotrienol) (RM3827 at 181 bp). A total of 28 plants were selected in BC<sub>1</sub>F<sub>1</sub> generation, which contained all desirable alleles for high amylose content, tocopherol and tocotrienol. On the basis of morphological characters, plant number 9 of BR16/2 × Karingam (BC<sub>1</sub>F<sub>1</sub>) was selected for high yield potential. This plant produced the highest grain weight per plant (10.7 g) among 28 selected plants with 77.2% filled grain and grain color of this plant was white which can be used for the development of good quality and high yield potential upland variety.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

**PENCIRIAN MORFOLOGI, NUTRISI, DAN MOLEKULAR UNTUK  
PENAMBAHBAIKAN GENOTIP PADI BUKIT MELALUI PEMBIAK-  
BAKAAN KACUKAN BALIK**

Oleh

**JANNATUL FERDOUS**

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Genotip padi bukit mempunyai hasil pengeluaran yang rendah tetapi menunjukkan trait-trait yang istimewa. Pemiakbakaan kacuk balik dibantu penanda terpilih merupakan teknik yang berguna dalam penambahbaikan genotip padi bukit dari aspek hasil dan kualiti, dimana trait-trait yang dikehendaki akan dipilih dalam proses awal penghasilan generasi kacuk balik. Tujuan kajian ini adalah untuk memasukkan kandungan alel-alel amilosa dan Vitamin E yang tinggi dalam generasi  $BC_1F_1$  serta memilih pokok dengan alel yang dikehendaki melalui marker assisted selection (MAS) dengan menggunakan penanda molekul jujukan ringkas terulang, SSR. Sebanyak 39 genotip padi telah dinilai untuk pemilihan pokok induk berdasarkan morfologi, kandungan nutrisi dan kepelbagaian genetik. Sebanyak tiga genotip padi bukit iaitu Bukit Garam582, Bukit Garam1449 dan Karingam telah dipilih dan dikacukan balik dengan dua varieti yang mempunyai hasil pengeluaran yang tinggi (BR16 dan MR219). Kaedah MAS digunakan untuk mengenalpasti pokok yang dikehendaki dengan kandungan amilosa dan Vitamin E dalam generasi  $BC_1F_1$ . Ciri-ciri variasi yang penting telah diperhatikan antara genotip yang menyumbang kepada hasil, kandungan klorofil, kandungan amilosa, indeks glyceemic, dan aktiviti

antioksidasi. Kepelbagaian 39 genotip padi telah dianalisis dengan meneliti 14 ciri-ciri morfologi. Tiga puluh enam genotip padi telah dikumpulkan kepada 3 kluster dan genotip Pulut Hitam Pahang(PHP), Lannsan, dan Karibang telah dikumpulkan kepada 3 kedudukan yang berbeza melalui analisis kluster morfologi. Empat komponen pertama dalam analisis komponen prinsipal (PCA) menerangkan variasi untuk semua ciri-ciri morfologi dengan kadar 76.5% dan menunjukkan kolerasi positif di antara bilangan anak, bilangan tangkai padi, peratusan biji terisi, dan bilangan biji per tangkai padi. Kandungan amilosa yang tinggi (27.1%), dan indeks glycemic yang rendah (71.1%) telah didapati pada BR16. Nilai indeks glycemic yang tertinggi (99.2%) pada Bukit Hitam (beras pulut) dan juga mempunyai kandungan amilosa yang rendah (1.5%). Indeks glycemic dan kandungan amilosa menunjukkan kolerasi negatif. Kandungan antioksidasi yang tinggi adalah pada Karingam (89.8%) dan paling rendah pada BR16 (50.8%). Kandungan Fe, Zn, dan Mn yang tinggi didapati pada genotip putih dan genotip yang tinggi dalam hasil pengeluaran. Kandungan Mn dan Cu yang tinggi telah dikesan pada genotip hitam tempatan (54.4  $\mu\text{g/g}$ ) dan genotip merah padi hijau manis (23.2  $\mu\text{g/g}$ ). Kepelbagaian genetik untuk 39 genotip telah dinilai dengan 25 penanda molekul SSR. Analisis kluster menunjukkan perbezaan genotip padi kepada 4 kumpulan utama dan beberapa subkumpulan. Sepuluh kombinasi kacukan telah dibuat daripada kacukan silang tunggal dan kacukan silang berganda diantara 5 induk yang mempunyai kepelbagaian genetik. Pokok  $F_1$  telah dibuktikan menggunakan dengan penanda Wx, RM190, RM3187, dan RM3827. Penanda Wx dan RM190 digunakan untuk memilih alel berlilin untuk kandungan amilosa yang tinggi. Penanda RM3187 dan RM 3827 telah digunakan untuk memilih alel HGGT dan HPT yang berperanan untuk biosintesis tocoferol dan tocotrienol. Penanda berikut diampifikasi: untuk



kandungan amilosa yang tinggi (RM190 dan Wx pada fragmen bersaiz 107 bp); tinggi aktiviti alel qTOC-6-2 (tocoferol) (RM3187 pada 148 bp) dan qT3-6-1 (tocotrienol) (RM3827 pada 181 bp). Sebanyak 28 tumbuhan telah dipilih daripada generasi BC<sub>1</sub>F<sub>1</sub> yang mempunyai semua kandungan alel-alel yang dikehendaki untuk kandungan amilosa, tocoferol, dan tocotrienol yang tinggi. Berdasarkan kepada ciri-ciri morfologi, pokok padi nombor 9 daripada BR16/2 × Karingam (BC<sub>1</sub>F<sub>1</sub>) telah dipilih untuk potensi tinggi hasil pengeluaran. Pokok ini memberikan berat biji per pokok yang tertinggi (10.7 g) daripada 28 pokok yang terpilih dengan 77.2% biji terisi dan warna biji pada tumbuhan ini adalah putih dan boleh digunakan untuk penambahbaikan varieti yang mempunyai kualiti yang baik dan hasil pengeluaran yang tinggi.

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I certify that an Examination Committee met on 23 April 2012 to conduct the final examination of Jannatul Ferdous on her Doctor of Philosophy thesis entitled “Morphological, nutritional, and molecular characterization of selected genotypes for upland rice improvement through marker-assisted backcross breeding” in accordance with Universiti Pertanian Malaysia (Higher Degree) Act 1980 and Universiti Pertanian Malaysia (Higher Degree) Regulation 1981. The Committee recommends that the candidate be awarded the relevant degree.

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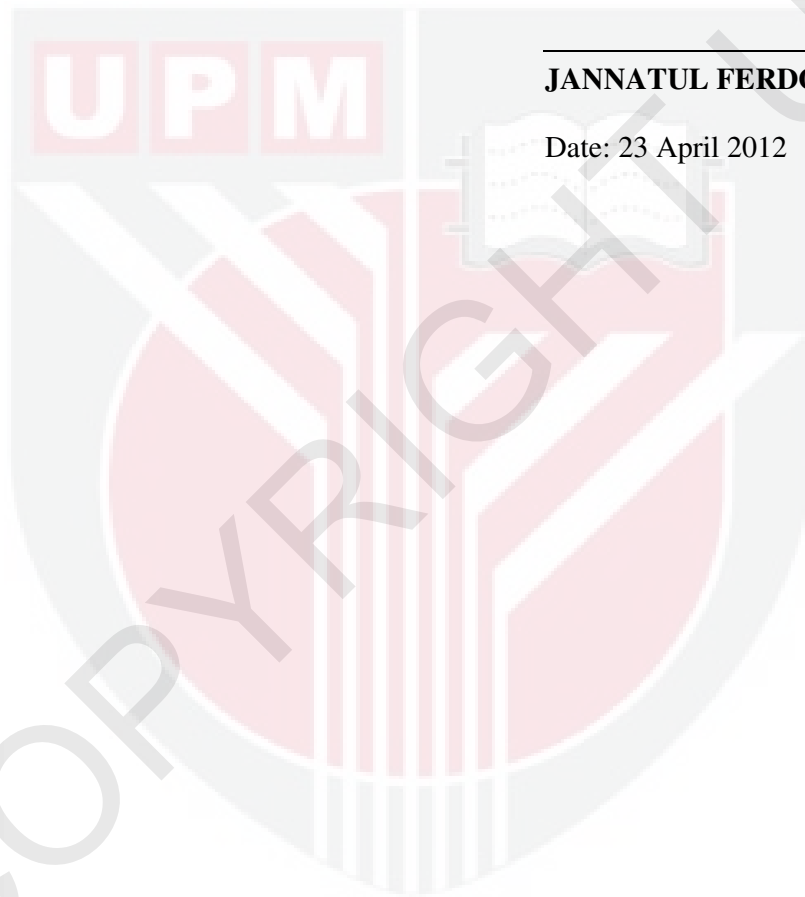
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## DECLARATION

I declare that the thesis is my original work except for equations 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 other institutions.



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