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HISTOLOGICAL AND BIOCHEMICAL CHARACTERIZATION OF OIL PALM (ELAEIS GUINEENSIS JACQ.) ENDOSPERM DURING FRUIT DEVELOPMENT

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By

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**Thesis Submitted to the School of Graduate Studies,
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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

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Chairman : Parameswari a/p Namasivayam, PhD

Faculty : Biotechnology and Biomolecular Sciences

Oil palm (*Elaeis guineensis*) is commercially planted in Malaysia and Indonesia for palm oil production. Palm oil has become the largest source of edible oil in the world. With increasing global demand for vegetable oil coupled with the challenges in production, there is an urgent need to improve palm oil productivity. Therefore, *in vitro* propagation of oil palm has been in progress to regenerate desirable traits in order to meet the world demand. However, the oil palm tissue culture technique is still inefficient and there is also no universal culture medium for all genotypes. One of the focus areas is the optimization of media formulation for enhanced somatic embryogenesis through the understanding of endosperm development. Thus, the aim of this study was to investigate changes in nutrient composition and morphology of endosperm during fruit development.

Histological features of oil palm fruits were examined by histochemistry and light microscopy. Oil palm endosperm formation started at 2 weeks after anthesis (WAA) and it was in liquid form. Enlargement of endosperm occurred as the seed developed

further. Cellularization occurred at 8 WAA and cells started grow around periphery of the endocarp. The cells in endosperm continued to grow towards the centre of endosperm. The mature endosperm contained numerous nutrient reserves which was essential for nurturing embryo growth.

Biochemical compositions in developing oil palm fruits were analysed by chromatographic and atomic spectroscopic techniques. Glutamic acid was the predominant amino acid followed by aspartic acid and arginine throughout the endosperm developmental stages of both planting materials, tenera and clonal. It was suggested that the glutamic acid, aspartic acids and arginine were the main building block in kernel protein. At early stages of seed development, the high hexose/sucrose ratio in tenera (57.85) and clonal (250.53) was observed which is important to meet the high metabolic demand required for cell division and differentiation. However, the ratio was switched at 10WAA in both tenera and clonal (0.80 and 1.53, respectively) which indicated the onset of endosperm maturation phase. Analysis of fatty acids revealed that lauric acid was the most abundant fatty acid in mature oil palm endosperm of tenera (54%) and clonal (51%).

The macro and micro mineral elements are essential for plant growth. High amount of manganese and calcium were found at 5 WAA. Manganese content found in tenera and clonal endosperm was 68 mg/100g dw and 158 mg/100g dw, respectively. While, calcium content found in tenera and clonal endosperm was 797 mg/100g dw and 6350 mg/100g dw, respectively. For both tenera and clonal, potassium (108 and 751 mg/100g dw, respectively) and phosphorus (653 and 695 mg/100g dw

respectively) were accumulated in mature endosperm. Analyses of vitamins showed that alpha-tocopherol and niacin were the predominant vitamins found in both oil palm endosperms.

On the whole, the anatomical and nutritional study of oil palm endosperm has provided information on cellular changes and nutrient requirements for zygotic embryo development. This knowledge will be useful in optimizing the tissue culture media of oil palm in order to improve the efficiency of somatic embryogenesis.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

PENCIRIAN SIFAT HISTOLOGI DAN BIOKIMIA ENDOSPERMA KELAPA SAWIT (*ELAEIS GUINENSIS* JACQ.) SEMASA PERKEMBANGAN BUAH

Oleh

KOK SAU YEE

Jun 2011

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Kelapa sawit (*Elaeis guineensis*) ditanam secara komersial di Malaysia dan Indonesia untuk pengeluaran minyak sawit. Minyak sawit telah menjadi sumber minyak masak yang terbesar di dunia. Pengeluaran minyak sawit yang bertepatan telah menjadi satu keperluan bersandaran dengan peningkatan permintaan dunia terhadap minyak sawit serta cabaran dalam proses penghasilannya. Oleh sebab itu, propagasi *in vitro* bagi kelapa sawit telah dijalankan bagi menghasilkan pokok kelapa sawit yang mempunyai cirri-ciri yang diingini bagi memenuhi permintaan dunia. Namun, teknik kultur tisu kelapa sawit masih lagi tidak cekap dan tiada media kultur umum yang bersesuaian untuk semua genotip. Salah satu bidang yang perlu diberi perhatian ialah pengoptimuman formulasi media demi meningkatkan embriogenesis somatik, yang mana ianya dapat dicapai melalui pemahaman tentang perkembangan endosperma. Oleh yang demikian, kajian ini dijalankan bertujuan untuk menyiasat perubahan komposisi zat makanan dan morfologi endosperma semasa proses perkembangan buah.

Kajian mengenai ciri-ciri histology buah kelapa sawit telah dijalankan dengan teknik histokimia dan mikroskopi cahaya. Pembentukan endosperma kelapa sawit bermula pada 2 minggu selepas antesis (‘weeks after anthesis’, WAA) di mana endosperma adalah dalam bentuk cecair. Pembesaran endosperma terus berlaku seiring dengan perkembangan biji benih. Pengeselan berlaku pada tahap 8 WAA dan pertumbuhan sel-sel mula dilihat pada bahagian bawah endokarp. Sel-sel di sekeliling endosperma berkembang ke arah bahagian tengah endosperma. Endosperma yang matang mengandungi banyak simpanan zat makanan yang sangat diperlukan untuk pertumbuhan embrio.

Kajian mengenai komposisi biokimia dalam buah kelapa sawit yang sedang berkembang telah dianalisis dengan teknik kromatografi dan spektroskopi atom. Dalam kedua-dua bahan kajian yang digunakan iaitu ‘tenera’ and ‘clonal’, asid glutamik merupakan asid amino yang dominan pada setiap tahap perkembangan endosperma dan diikuti dengan asid aspartik dan arginina. Ini menunjukkan bahawa asid glutamik, asid aspartik dan arginina ialah blok binaan utama dalam protin isirong. Pada tahap awal perkembangan biji benih, nisbah heksosa/sukrosa yang tinggi dalam ‘tenera’ (57.85) dan ‘clonal’ (250.23) telah dikenalpasti di mana ianya adalah penting bagi memenuhi keperluan metabolik yang tinggi untuk proses pembahagian dan pembezaan sel. Namun, nisbah tersebut telah berubah pada tahap 10 WAA bagi kedua-dua ‘tenera’ dan ‘clonal’ (0.80 dan 1.53, masing-masing), dan ini menandakan bermulanya fasa pematangan endosperma. Analisis asid lemak pula menunjukkan bahawa asid laurik adalah asid lemak yang paling banyak dijumpai

dalam endosperma kelapa sawit yang matang bagi kedua-dua ‘tenera’ (54%) dan ‘clonal’ (51%).

Unsur-unsur mineral mikro dan makro adalah penting untuk pertumbuhan pokok. Jumlah kalsium dan mangan yang tinggi telah didapati pada tahap 5 WAA. Kandungan mangan dalam endosperma ‘tenera’ dan ‘clonal’ masing-masing ialah 68 mg/100g berat kering dan 158 mg/100g berat kering. Manakala, kandungan kalsium dalam endosperma ‘tenera’ dan ‘clonal’ ialah 797 mg/100g berat kering dan 6350 mg/100g berat kering. Bagi kedua-dua ‘tenera’ dan ‘clonal’, kandungan kalium (108 dan 751 mg/100g berat kering, masing-masing) dan fosforus (653 dan 695 mg/100g berat kering, masing-masing) telah berkumpul dalam endosperma yang matang. Analisis vitamin pula menunjukkan bahawa alfa-tokoferol dan niasin adalah vitamin utama yang ditemui dalam kedua-dua endosperma kelapa sawit.

Secara keseluruhannya, kajian anatomi and zat makanan endosperma kelapa sawit telah membekalkan maklumat mengenai perubahan selular dan keperluan nutrien untuk perkembangan embrio zigotik. Pengetahuan ini amatlah berguna dalam pengoptimuman media kultur tisu kelapa sawit supaya kecekapan embriogenesis somatik dapat ditingkatkan.

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I certify that a Thesis Examination Committee has met on 14 June 2011 to conduct the final examination of Kok Sau Yee on her thesis entitled "Histological and Biochemical Characterization of Oil Palm (*Elaies guineensis* Jacq.) Endosperm During Fruit Development" in accordance with Universities and University College Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The committee recommends that the candidate be awarded Master of Sciencce.

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

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I declare that the thesis is my original work except for quotations 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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KOK SAU YEE

Date: 14 June 2011

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