

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

**DIFFERENTIAL GENE EXPRESSION OF OIL PALM (*ELAEIS GUINEENSIS* JACQ.) FRUITS DURING LATE RIPENING STAGES AND MOLECULAR CHARACTERISATION OF METALLOTHIONEIN-LIKE TRANSCRIPTS**

By

**AHMED BAKHIT SALIM AL-SHANFARI**

**September 2012**

**Chairman: Associate Professor Datin Siti Nor Akmar binti Abdullah, PhD**

**Faculty: Agriculture**

Oil palm (*Elaeis guineensis* Jacques) is a major oil crop of economic importance in the world. Different biological and physiological mechanisms occur during oil palm fruit ripening. These mechanisms are regulated by the genes expressed during fruit ripening processes. Therefore, determining the structure and function of the expressed genes involved in these mechanisms will help to improve the important characters for enhancing palm fruit oil yield and quality. A forward suppression subtractive hybridization (SSH) library was constructed to identify genes involved in fruit ripening in oil palm. Oil deposition, as an oil palm fruit ripening indicator, will start at approximately 12 weeks after anthesis (w.a.a.) with an active period of oil synthesis at 15-16 w.a.a. until fruit maturity at about 20 w.a.a. in the mesocarp. Hence, the suppression was performed with cDNA extracted from fruits at 12 w.a.a. as “driver” and fruits at 17 w.a.a. as “tester”. A single-pass sequencing from the 5'-end of 2,112 randomly selected cDNA clones resulted in 2,019 high-quality expressed sequence tags (ESTs). Clustering of the 2,019 EST sequences showed 20 unigenes consisting of 9 contigs and 11 singletons. Among the edited EST sequences, 1,109 (14 unigenes) had significant matches with protein sequences in the public

databases. The matched ESTs were further classified into six putative cellular functions including unclassified proteins (38.0%), cell rescue and defense (33.8%), proteins with binding functions (27.7%), biogenesis of cellular component (0.3%), metabolism (0.1%), and cell cycle (0.1%). At 17 w.a.a., the 20 unigenes were expressed at higher abundance compared to 12 w.a.a.

Two of the abundant transcripts encoding type 2 metallothionein-like proteins designated as *MET2a* and *MET2b* were selected for further study due to their high abundance (16.05%) in the SSH library and their involvement in biological processes in fruit development and maturation. The second part of the present study involved the isolation of the full-length cDNA encoding *MET2a* and *MET2b* from the ripening oil palm fruit mesocarp at 17 w.a.a. and examining their expression patterns compared to the other two previously reported type-3 MT members (*MT3-A* and *MT3-B*) in various oil palm organs including different vegetative and reproductive tissues. The full-length cDNA sequence of *MET2a* and *MET2b* were 571 and 553 bp designated as *EgMT2a* and *EgMT2b*, respectively. Their sequences were homologous (67–77%) with several type-2 MTs in plants. All four genes were differentially expressed in the ripening oil palm mesocarp tissues but undetectable in the vegetative tissues examined. All the MT genes examined were significantly up-regulated in the mature developmental stages of oil palm fruit mesocarp except for *EgMT2b* which was expressed only at 17 w.a.a. but at a much lower level. The type 2 MT-like proteins are more in relation with late fruit ripening stage than the type 3 MT-like proteins consistent with their reported functions in the homeostasis or detoxification. The findings in the present study will contribute to a better understanding of the molecular mechanisms involved in fruit ripening in oil palm.