

BETA-KETOACYL ACP SYNTHASE II (KAS II)

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Novel palm oil compositions are needed to meet the demand of the expanding oil business and changing market requirements. Genetic engineering provides the opportunity to diversify the use and improve the economic value of palm oil. Reported successes in changing fatty acid compositions in other crops provide examples of how *in vitro* gene technology can be used as a means for modifying both fatty acid chain length and level of fatty acid unsaturation. Changing oil compositions by *in vitro* gene technology requires tools and techniques such as isolation of the genes of interest, plant regeneration system, a reliable transformation technique for ensuring stable integration and regulatory sequence for controlling expression of introduced genes (Figure 1). In oil palm, the focus of the genetic engineering programme is to develop new varieties which have different oil characteristics such as high oleate. It is envisaged that higher oleate content will serve as a feedstock for the oleochemical industry and facilitate the entry of palm oil into the liquid oil market.

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THE IMPORTANCE OF KAS II GENE

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There is strong evidence that KAS II is an important regulatory enzyme that is responsible for palmitic acid accumulation in the oil palm mesocarp. Increasing the activity of the KAS II will decrease the palmitic acid and increase oleic acid content of palm oil (Figure 2). Thus, efforts at manipulating fatty acid composition of palm oil require the full-length cDNA of the KAS II. Additionally, coordinate expression of the KAS II gene with other enzymes related to fatty acid biosynthesis is also desirable for the production of other higher value products that can benefit the industry in achieving higher profitability when compared with the production of commodity oil.

CHARACTERISTICS OF OIL PALM KAS II GENE

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A 2.0 kb full-length KAS II cDNA was isolated from 17-week oil palm (*Elaeis guineensis*) mesocarp (Figure 3). The full coding sequence of this gene is being used in the genetic engineering programme to increase the oleic acid content of palm oil. Sequence analysis revealed that the KAS II sequence contains 1650 base pairs (bp) of open reading frame coding for 549 amino acids. The oil palm KAS II cDNA shows very good sequence homology to other plant KAS II cDNAs in the Genbank database. Northern blot analysis showed that expression of KAS II in the mesocarp was low in younger tissues and higher at 17- and 20-week stage, which correlates well with the period of palm oil synthesis (Figure 4). High expression was also observed in 12-week kernel and germinated seedlings but very negligible in roots and young leaves.



Figure 1. Production of novel product in oil palm mesocarp.

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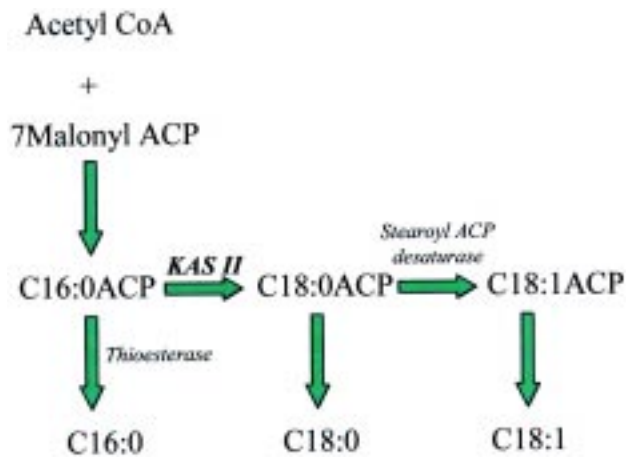


Figure 2. Fatty acid biosynthesis pathway.

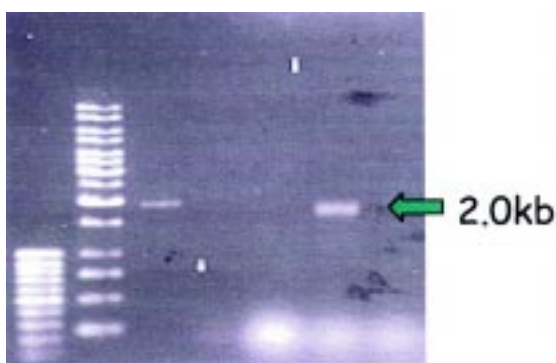
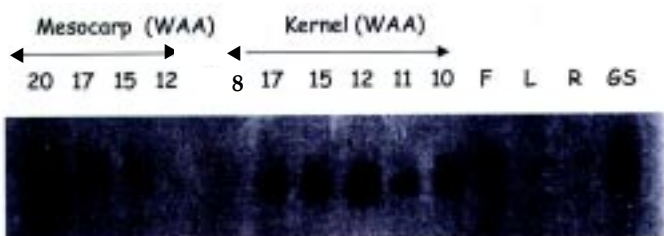


Figure 3. Full length KAS II from *Elaeis guineensis*.



WAA = Week after anthesis; F = Flower; L = Leaf; R = Root; GS = Germinated seedlings

Figure 4. Expression of KAS II in oil palm tissues.

FUNCTION STUDIES OF KAS II

Two model species namely *E. coli* and *Arabidopsis thaliana* have been employed to examine the consequence of over expressing or down regulating KAS II expression and to discover that data from these systems support the hypothesis that this may be a key enzyme in the modulation of oil quality. Analysis of *E. coli* cells containing the full-length KAS II cDNA construct and control cells lacking this

construct was conducted to determine their fatty acid compositions. Results demonstrate that the percentage of C18 fatty acids in *E. coli* increases at the expense of C16:0 upon expression of the oil palm KAS II protein (Figure 5). In another experiment, an antisense KAS II construct was used to transform *Arabidopsis thaliana*. Fatty acid analyses of transformed and control seeds were performed (Figure 5). Higher palmitic acid was detected in the transformed plants compared to control, thus being in agreement with our postulation that KAS II is

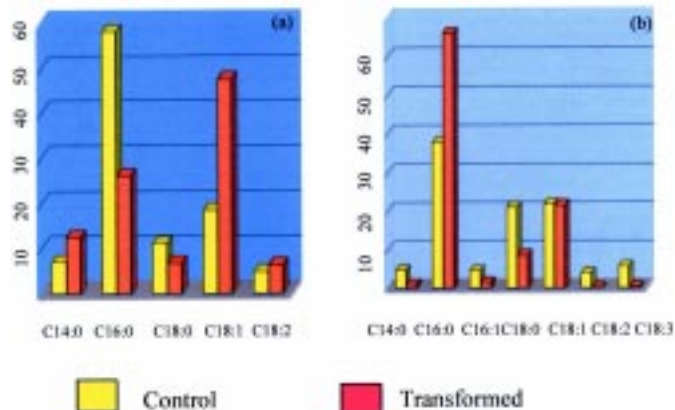


Figure 5. Functional and expression studies on KAS II in (a) *E. coli* (b) *Arabidopsis thaliana*.

important in controlling palmitic acid accumulation of palm oil. □

CONCLUSION

□ A full-length KAS II cDNA was isolated from the oil palm (*Elaeis guineensis*). The KAS II was proven to be functional. The full-length cDNA is available for licensing to interested parties for genetic modification of oil crops.

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