PHYTOENE SYNTHASE GENE FROM OIL PALM (*Elaeis oleifera*) FOR MODIFICATION OF CAROTENOID CONTENT

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Malaysia is the second largest producer of palm oil in the world. The major problems faced by the industry are labour and arable land shortages. MPOB has identified genetic engineering as a promising strategy that could be utilised to face the above challenges (Parveez, 1998). As shown in other crops, palm oil carotene composition can also potentially be modified by manipulating the activity of the key enzyme involved in carotenoid biosynthesis namely, phytoene synthase (PSY). There are two species of oil palm, *Elaeis guineensis* and *Elaeis oleifera*. *E. oleifera* produces oil with a higher amount of total carotene content of about 4000 parts per million (ppm) compared to *E. guineensis* (500-700 ppm) (Yap *et al.*, 1991). Therefore, it is believed that *E. oleifera*'s *psy* is more active than *E. guineensis*' *psy*. As such, carotenoid content of *E. guineensis* could potentially be increased by overexpressing *psy* gene from *E. oleifera*.

ISOLATION OF PHYTOENE SYNTHASE GENE FROM *Elaeis oleifera*

PSY is one of the key enzymes involved in carotenoid biosynthesis. It catalyses the head-to-head condensation of two geranylgeranyl diphosphate molecules to produce colourless phytoene. A full-length cDNA encoding PSY (designated as PSYEO) was isolated from the mesocarp of *E. oleifera* (*Figure 1*). The cDNA contains a 1298-bp open reading frame which encodes 432 amino acid residues. The deduced amino acid sequence of PSYEO shares approximately 98% and 80% sequence identity with PSY from *E. guineensis* and other plants (*Figure* 2), respectively. Results from Southern blot analysis indicate that *E. oleifera* has a single copy of *psy* gene (*Figure 3*).

BENEFITS OF PHYTOENE SYNTHASE GENE

E. oleifera has been shown to contain much higher levels of carotenoids, up to 4000 ppm (Choo *et al.*, 1995) compared to *E. guineensis*. In the plant carotenoid biosynthetic pathway, PSY catalyses the first commited step and is considered as a rate limiting enzyme. Up regulating *psy* has been shown to increase total carotenoid in plants, including tomato (Fraser *et al.*, 2002) and rapeseed (Shewmaker *et al.*, 1999). Therefore, introduction of *psy* from *E. oleifera* to oil palm (*E. guineensis*) could potentially increase the carotenoid accumulation in transgenic oil palm. Moreover, this gene can also be used to increase carotenoid content in other crops such as rice, tomato and pepper. Currently, according to 2011 report by Business Communication Company in *The Global Market for Carotenoids*

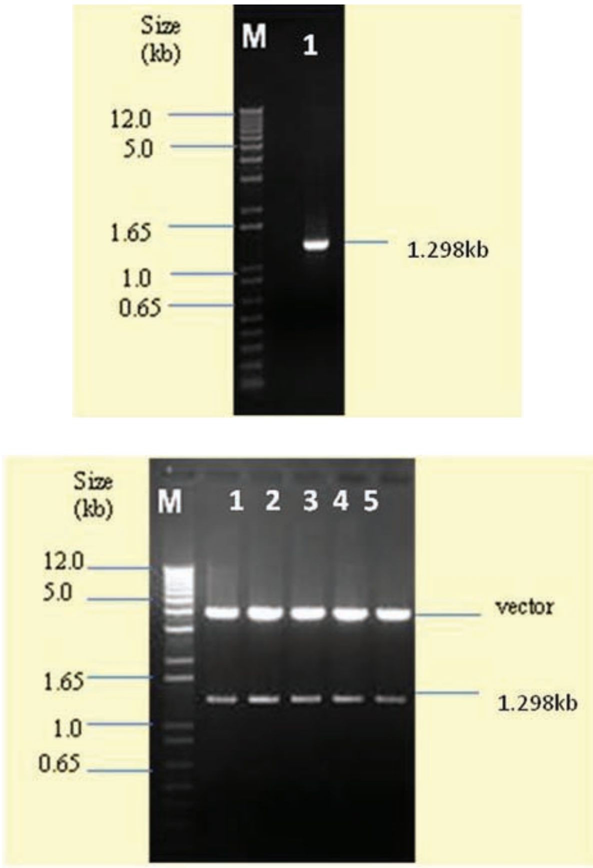

Figure 1. A: Amplification of the full-length cDNA region for E. oleifera psy *using PSYA and PSYB primers. Arrow shows the 1.298 kb amplified product. B: Clones containing amplified product from cDNA. Lane M: 1 KB plus DNA.*

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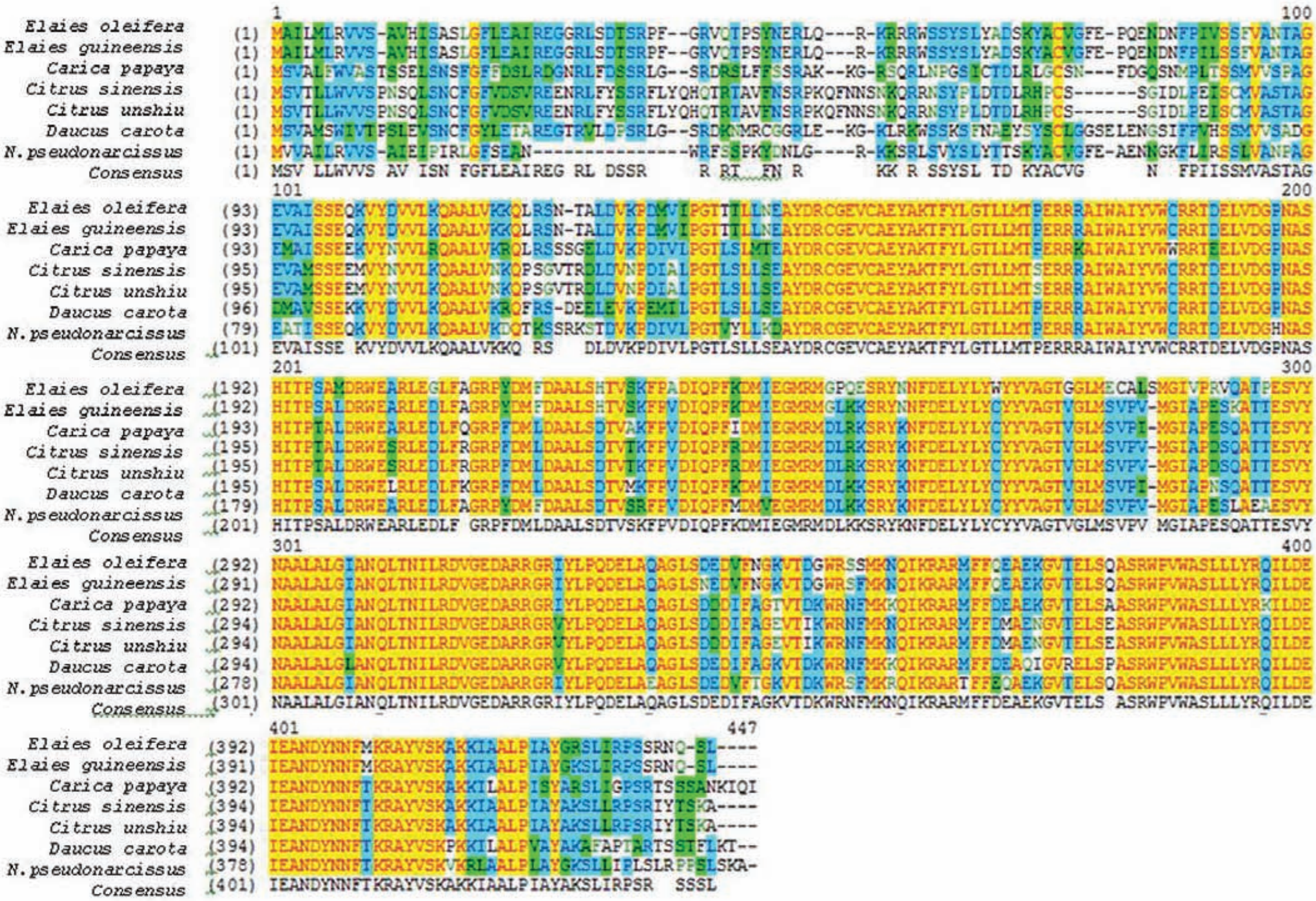

Figure 2. Alignment of the amino acid sequence of phytoene synthase from E. oleifera *with other plant phytoene synthases. Conserved amino acids are indicated by yellow colour.*

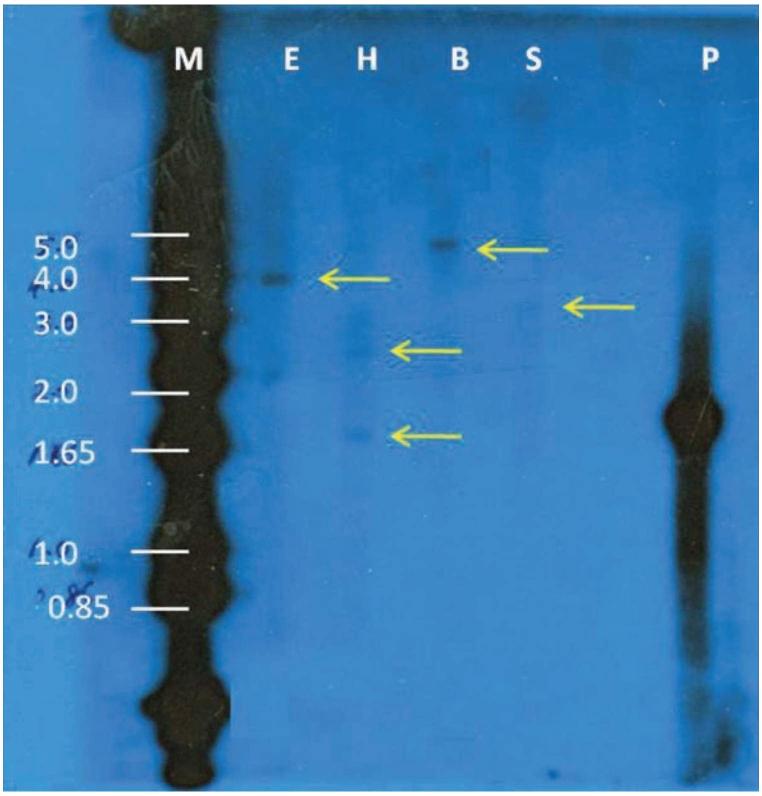

Figure 3. Southern blot analysis of E. oleifera psy*. Thirty microgrammes of total DNA from* E. oleifera *were digested using four different restriction enzymes namely* EcoR I, Hind III, BamH I *and* Spe I*. M, 1kb plus DNA ladder; E,* EcoR I*; H,* Hind III*; B,* BamH I*; S,* Spe I*; P, plasmid. Arrows show the location of bands.*

(http://www.bccresearch.com), the carotenoid market is predicted to grow to RM 1.4 billion in 2018 with a compound annual growth rate of 2.3%. The demand for carotenoids is increasing because of growing evidence that enhancing specific carotenoid content in the diet, may help prevent or control particular diseases and disorders. For example, consumption of lycopene has been shown to lead to a decreased risk of prostate cancer (Dahan *et al.*, 2008).

WHO WOULD BENEFIT?

Molecular biologists or biotechnologists from the oil palm industry can benefit from using the cDNA to manipulate the carotenoid content. Similarly, molecular biologists and biotechnologists from local universities, research institutions and research-based companies can benefit from the use of the cDNA in heterologous systems such as bacteria or other plants.

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