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Astaxanthin biosynthesis in transgenic *Arabidopsis* by using *Chyb* gene encoding – carotene hydroxylase

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Objectives

We have tried to isolate *Chyb* gene encoding -Carotene Hydroxylase, construct Ti-plasmid vector and conduct transformation to *Arabidopsis*. Transgenic plants were investigated molecular characterization and chemical analysis.

Materials and Methods

- 1. Materials: Transgenic Arabidopsis plants
- 2. Methods: Breeding of transgenic plants using *Agrobacterium tumefacience*, Molecular characterization (Southern blot, RT-PCR analysis, RealTime PCR, Northern blot) and Biochemical analysis: HPLC analysis

Results and Discussion

Oxycarotenoids are oxygenated carotenoids that perform critical roles in plants. -Carotene hydroxylase adds hydroxyl groups to the -rings of carotenes and has been cloned from several bacteria and plants including *Arabidopsis*. This study was carried out to investigate the effect of -carotene hydroxylase gene (*Chyb*) on the oxycarotenoids biosynthesis in the transgenic *Arabidopsis*. Construct of pGCHYB containing *Chyb* was established onto Gateway vector system (pENTR3C gateway vector and pH2GW7 destination vector). *Arabidopsis thaliana* (cv. Columbia) was transformed with *Agrobacterium tumerfacience* GV3101 harboring pGCHYB construct driven by 35S promoter and hygromycin resistant gene. Seven hundred bases paired PCR products, indicating the presence of *Chyb* gene, were found in the transformants by PCR analysis using *Chyb* primers. Hygromycin resistance assay showed that transgenes were stably inherited to next generation. The overexpression of the *Chyb* gene resulted in the decrease carotenoid content. Especially, astaxanthin unusual oxycarotenoid in wild type *Arabidopsis* was detected in the transgenic plants. This means that decreased carotenoids might be converted into astaxanthin metabolism with the aid of silent gene in the host.

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