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Carotenoid content and expression of phytoene synthase and phytoene desaturase genes in bitter melon (*Momordicacharantia*)

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Objectives

In this study, the relationship between carotenoid accumulation and the gene expression of *PSY* and *PDS* was investigated in different organs of and stages of fruit ripening in *M. charantia*. In addition, principal component analysis (PCA) was used to evaluate the differences among the profiles of 7 carotenoids identified in its fruit at several maturation stages.

Materials and Methods

- Plant Material: *M. charantia* seeds were purchased from Asian Seed Company (Seoul, Korea) and stored at 4 °C. *M. charantia* seeds were germinated in a greenhouse, and then the seedlings were transferred to the experimental farm at Chungnam National University (Daejeon, Korea).
- Real-Time Polymerase Chain Reaction Analysis
- Extraction and High Performance Liquid Chromatography Analysis of Carotenoids
- Principal Component Analysis.

Results

Momordica charantia Linn, which belongs to the Cucurbitaceae family, is a medicinal vegetable that grows in the tropical and subtropical parts of the world. It is commonly known as “bitter melon” or “bitter gourd” because all parts of the plant, including the fruit, taste bitter. In traditional medicine, bitter melon has been used as a remedy for diabetes. This plant, produces a fruit that has a β-carotene concentration 5 times higher than that of carrot. To elucidate the molecular basis of β-carotene accumulation in *M. charantia*, the gene expression levels of phytoene synthase (*McPSY*) and phytoene desaturase (*McPDS*) were determined. These levels were particularly high in the flowers of *M. charantia*. During fruit maturation, the expression levels of *McPSY* and *McPDS* decreased during the mid stages but increased in the fully mature fruit. In addition, carotenoids accumulated as the peel changed from green to orange. Thus, *McPSY* and *McPDS* expression correlated with carotenoid accumulation during fruit maturation. Principal component analysis (PCA) also was used to evaluate the differences among the profiles of 7 carotenoids identified in the fruit at several maturation stages. Riper fruits had higher carotenoid concentrations than less ripe fruits.

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Table 1. Carotenoid content in different organs of *M. charantia* (μg $\times g^{-1}$dry weight). Results are expressed as mean (SE) (n=3). N.D., not detected; O-leaf, oldleaf; Y-leaf, youngleaf; M-flowers, male flowers; F-flowers, female flowers.

	roots	stems	O-leaves	Y-leaves	M-flowers	F-flowers
lycopene	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
α-carotene	0.6 (0.2)	3.3 (0.2)	23.2 (2.4)	10.2 (0.8)	7.0 (2.0)	7.8 (0.6)
lutein	10.4 (5.3)	110.8 (5.3)	284.3 (91.8)	223.9 (49.2)	98.1 (7.1)	120.1 (21.7)
β-carotene	23.5 (13.4)	262.4 (8.8)	743.8 (272.5)	634.2 (192.7)	394.8 (8.6)	356.4 (34.6)
β-cryptoxanthin	0.6 (0.3)	1.0 (0.2)	3.5 (0.3)	3.5 (0.8)	6.7 (0.9)	7.6 (0.4)
zeaxanthin	0.0 (0.0)	0.1 (0.0)	0.5 (0.0)	0.6 (0.1)	0.3 (0.0)	0.3 (0.0)
antheraxanthin	0.4 (0.1)	2.9 (0.2)	7.5 (0.4)	9.2 (1.3)	15.1 (0.5)	5.3 (0.4)
violaxanthin	0.9 (0.3)	8.5 (0.7)	40.3 (3.0)	29.6 (4.5)	45.1 ()	14.8 (0.5)

Table 2. Carotenoid content during different stages of fruit maturation in *M. charantia* (μg $\times g^{-1}$dry weight). Results are expressed as mean (SE). N.D., not detected.

	Stage 1	Stage 2	Stage 3	Stage 4	Stage 5	Stage 6
lycopene	N.D.	N.D.	N.D.	N.D.	N.D.	14.6 (2.3)
α-carotene	14.6 (1.3)	10.9 (0.8)	6.5 (0.1)	4.5 (0.5)	8.0 (0.3)	4.5 (1.1)
lutein	122.2 (5.9)	85.5 (7.2)	52.8 (2.6)	46.2 (4.9)	61.8 (1.8)	19.0 (6.0)
β-carotene	274.5 (30.3)	189.4 (11.8)	91.4 (4.2)	73.2 (8.2)	124.3 (8.5)	332.4 (72.1)
β-cryptoxanthin	2.4 (1.6)	1.0 (0.1)	0.5 (0.2)	0.4 (0.2)	0.5 (0.2)	174.3 (44.0)
zeaxanthin	0.2 (0.0)	0.1 (0.0)	0.1 (0.0)	0.1 (0.0)	0.1 (0.0)	2.2 (0.7)
antheraxanthin	3.0 (0.3)	2.1 (0.2)	0.6 (0.1)	0.6 (0.1)	0.9 (0.1)	8.8 (3.0)
violaxanthin	7.8 (1.4)	5.4 (0.4)	2.8 (1.1)	1.6 (0.2)	1.7 (0.1)	3.0 (1.0)

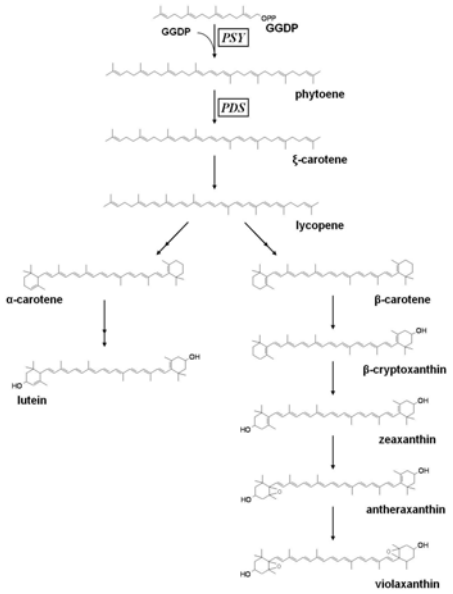

Fig. 1. Carotenoid biosynthesis pathway in plants. PSY, phytoene synthase; PDS, phytoene desaturase; GGDP, geranylgeranyl diphosphate.

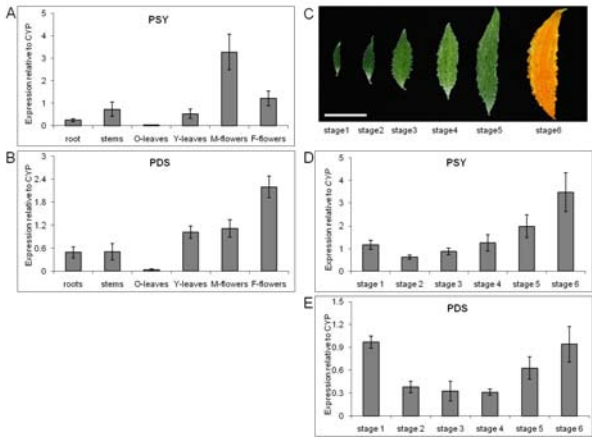

Fig. 2. Gene expression analysis of *PSY* and *PDS* in different organs and during fruit maturation in *M. charantia*.(A,B) Expression of *PSY* and *PDS* in different organs. (C) Photograph of the 6 developmental stages of *M. charantia* fruit. Fruit stages were determined by size.(D,E) Expression of *PSY* and *PDS* during fruit maturation. The height of each bar and the error bars show the mean and standard error, respectively, from 3 independent measurements. O-leaves, oldleaves; Y-leaves, youngleaves; M-flowers, maleflowers; F-flowers, femaleflowers; CYP, *M. charantia* cyclophilin.

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