

INCREASING β -CAROTENE CONTENT IN CASSAVA ROOTS BY EXPRESSION OF BACTERIAL GENES OF THE CAROTENOID PATHWAY

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A current strategy to improve the nutritional quality of cassava storage roots is the over expression of carotenogenic genes in a tissue specific manner. In transgenic plants, the bacterial Phytoene Synthase *crtB* gene catalyzes the conversion of Geranyl-Geranyl Diphosphate (GGPP) to phytoene in the first step of carotenoid biosynthesis. This study aimed to develop the cassava transgenic plants using the *crtB* gene fused to the CP1+ promoter. The promoter was isolated from a Glutamic Acid-Rich Protein (GARP) gene from cassava roots. The analysis of gene expression in roots of eight transgenic lines demonstrated that the promoter CP1+ was effective in expressing the *crtB* gene in storage roots of six-month old, field-grown plants. Transgenic cassava line 12B proved that it is possible to increase the total carotene content in storage roots up to 41 times. The amount of β -carotene also increased 17 times in the same line respect to the control, and a significant proportion (39%) of the carotenes were uncolored ones, like phytoene, phytofluene and β -carotene. This fact suggests that, to increase even more the β -carotene concentration in roots, it would be desirable to add at least two more genes of the bacterial pathway to cassava, *crtI* and *crtY* (Phytoene Desaturase and Lycopene \square -Cyclase), therefore we are introducing a carotene synthesis mini-pathway of three genes (*crtB*, *crtY* and *crtI*) from *E. urodevora* into cassava. These genes are under the control of root specific promoters isolated from cassava, sugar beet, potato and yams. Based on evidence from other crops like potato, it is expected that the expression of this pathway, which converts GGPP into β -carotene, may turn out to be a more efficient way of increasing the pro-VitA content in cassava than introducing just one *crtB* gene. Plants with three genes have been regenerated, confirmed transgenic via real time RT-PCR and Southern Blot, and soon will be harvest to check for carotenoid content after six months of growth in the field.

Keywords: *Manihot esculenta*, biosynthesis carotenoids, nutritional breeding.

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