Amylose-free sweet potato developed by RNAi of *GBSSI*

T. Shimada[1*], M. Otani[1], T. Hamada[1], S-H. Kim[1],
Y. Takahata[2], K. Katayama[2], K. Kitahara[3] and T. Suganuma[3]
[1]Ishikawa Agri College, Nonoichi, Ishikawa, 921-8836, Japan
[2] Nat. Agr. Res. Cent. Kyushu and Okinawa Region, Miyakonojyo, Miyazaki, 885-0091, Japan
[3] Kagoshima University, 1-21-24 Korimoto, Kagoshima, Japan

Objectives

Sweet potato (*Ipomoea batatas* (L.) Lam.) provides staple food and an important industrial raw material for human beings. Starch is the major component of storage roots of sweet potato. Genetic engineering of starch has a great potential for the quality improvement of sweet potato to meet the requirements of many established dietary and industrial applications and for the development of new dietary and industrial products. GBSSI is one of the key enzymes in the biosynthesis of starch, which catalyzes the formation of amylose in starch granules. By RNAi of *GBSSI* we have developed sweet potato plants containing novel starch, amylose-free starch.

Materials and Methods

The plasmid containing the sense orientation of *GBSSI* first exon , first intron and the antisense orientation of the first exon driven by CaMV35S promoter was introduced into embryogenic calli of sweet potato cultivar Kokei 14 through the *Agrobacterium tumefacience*-mediated transformation. The hygromycin resistant plants grew in a biohazard greenhouse for four months and their morphology and yield were investigated. Starches isolated from the root tubers were examined for amylose content and the physicochemical properties.

Results and Discussion

No morphological differences were observed between non-transgenic plants and the transgenic plants. Sliced roots of 28 of the 38 transgenic lines showed red-brown staining by iodine, while the remaining 10 lines showed dark blue staining similar to non-transgenic plants. The starches from red-brown stained roots contained no amylose calculated by the blue value at 680 nm. The yield and the starch content of the amylose-free lines exhibited no differences from those of non-transgenic plants and transgenic lines having a normal amylose content.

The transgenes were confirmed in all lines of transgenic plants by Southern hybridization. The results of Northern analysis revealed the amylose-free lines did not express the *GBSSI* mRNA. The

*Correspondig author : Takiko Shimada, Tel : 81-76-248-3135, E-mail : shimada@ishikawa-c.ac.jp

analysis by SDS-PAGE showed that they did not produce the 60 kD GBSSI protein. The results of the study showed that the double stranded RNA inhibits the expression of *GBSSI* gene completely in 73.7 % of the transgenic plant lines. The RNA interference is effective technology for the genetic improvement of crops.

The physicochemical properties of starches from amylose-free transgenic plants, transgenic plants with normal amylose content and non-transgenic plants were analyzed. The amylose-free transgenic plants appeared to contain starch with resistance to retrogradation, and different distribution of amylopectin chain length, compared to control. These unique characteristics may be useful for special products within the food industry.

TAKIKO SHIMADA

Present Address : 1-4-14 Togashi, Kanazawa, Ishikawa 921-8191, JAPAN

Education

1964 Bachelor of Agriculture, Kyoto University
1966 Master of Agriculture, Kyoto University
1972 Doctor of Agriculture, Kyoto University

Professional Experience

1984-1989 Associate Professor, Research Institute of Agricultural Resources, Ishikawa Agricultural College, Ishikawa, Japan
1989- Professor, Laboratory of Plant Cell Breeding, Research Institute of Agricultural Resources, Ishikawa Agricultural College, Ishikawa, Japan

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**Correspondig author* : Takiko Shimada, Tel : 81-76-248-3135, E-mail : shimada@ishikawa-c.ac.jp