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Studies on the expression of exogenous p5CS gene in transgenic wheatgrasses

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Key words : Wheatgrasses , Transgenic , p5CS gene , Northern blot

Introduction Transgenic wheatgrasses generated from hybrid wheatgrasses ($A gropyron cristatum \times A$. desertorum cv. Hycrest-Mengnong) were indentified by PCR analysis and Southern blot. The p5CS gene, which regulates the last step of proline synthesis in plants, was transferred into wheatgrasses. Northern blot was investigated. Results of Northern blot assay displayed that exogenous gene p5CS was expressed at transcription level in transgenic plants. New salt resistance plant lines which are adapted to extensive arid and semiarid areas of west China are expected to breed through these processes.

Materials and methods Plants of Hycrest-Mengnong wheatgrasses with p5CS gene that have been tested by PCR and southern blot were used as materials and compared to non transgenic plants. Plant total RNA was extracted by kit ; after plasmid DNA was amplified by PCR, arm fragments were extracted by kit as templates. They were labeled with DIG High prime DNA Labeling and Detection Starter Kit I by the random primer method ; electrophoresed RNA in formol denatured gel ; transferred using capillary blotting ; hybridized and detected by probe labeled with DIG.

Results Figure 1 shows the results of Northern blotting of p5CS transgenic plants and negative plants. The hybridization band of p5CS transgenic plants tested by PCR and Southern blot hybridized with the DIG probe was obvious. It proved that exogenous gene p5CS is expressed at transcription level in transgenic plants.



Figure 1 5 Northern blotting of transgenic wheatgrass plant . $1 \sim 5-$ transgenic plants ,6- negative control

Conclusions The p5CS catalyse the proline biosynthesis . Its activity , inhibited by proline content , p5CSF129A is a mutant wipe off feedback inhibition of p5CS and it led to a multiple increase of proline content , thus can enhance the protection of plant under osmotic stress . Now it has been transformed into many plant species as tobacco , rice , ryegrass and tall fescue to enhance the resistance to drought and salt . The results indicated that the exogenous gene p5CS was expressed at transcription level in transgenic plants .

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