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Molecular cloning and characterization of nitrate transporter genes from rice

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Objectives

1. Investigate extensive analyses of transcriptional regulation of OsNRT genes expression in response to nitrate starvation.
2. Determine the molecular mechanism by which rice plant respond to nitrate depletion in soil.
3. Generate new transgenic rice plants that can overcome the nitrate depletion in soil.

Meterials and Method

1. Material : Rice (Oryza sativa) cv. 'Dong-jin'
2. Method : Northern blot analysis, genomic/cDNA library screening, PCR cloning

Result and Discussion

We isolated the 3 different nitrate genes (OsNRT1-3) from rice (Oryza sativa). The encoded polypeptides are 82% identical to other plants and show high degree of amino acid sequence similarity with nitrate transporter gene of Arabidopsis thaliana, tobacco, soybean and barely. The OsNRT1 and OsNRT2 polypeptides are integral membrane proteins predicted to contain 12 membrane-spanning domains separated into two groups of six by a large charged hydrophilic region. OsNRT1 is 1601-bp long and contains an open reading frame encoding a 533 amino acid polypeptide, whereas OsNRT2 is 1550-bp long and encodes a 516 amino acid polypeptide. The two clones are 71% similar in their nucleotide sequence within the coding region. The two polypeptides are 56% identical in their amino acid sequence. The RNA blot analysis showed that expression of OsNRTs are various in response to nitrate deficiency. In particular expression of OsNRT1 and OsNRT2 were up-regulated in nitrate deficiency condition. However, OsNRT3 constitutively expressed in the both nitrate deficient and nitrate sufficient condition. Now we are generating trangenic rice plant overexpressing each OsNRT1 and OsNRT2 genes.

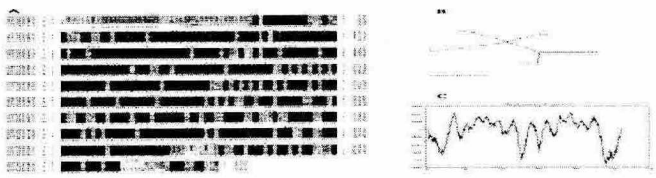

Fig. 1. Rice nitrate transporter (OsNRT1).

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