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# Screening of Rice (*Oryza sativa* L.) genotypes with response to salinity by RAPD marker

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Four genotypes of rice (Dandi, CSR-1, IR-36 and GR-3), differing in salt tolerance were grown at 3 and 5 EC ($dSm^{-1}$) salinity to study the effect of salinity at seedling (15 DAG) stage. The RAPD study indicated total 50.97% polymorphism and the maximum polymorphic *loci* was obtained by OP-D-8. Dandi at 3 EC and 5 EC recorded 100 per cent band sharing whereas the maximum genetic distance was recorded between CSR-1 (control) and GR-3 (5 EC) salinity. The polymorphisms in RAPDs are useful to study gene expression due to salinity in different rice cultivars.

Key words : RAPD, Rice, Salinity.

## INTRODUCTION

The complex stress-induced changes in physiology and growth of the plants are often the result of altered patterns of gene expression. Molecular markers based on the DNA sequence are more varied and reliable which can be used to identify and map the genes affecting complex plant traits such as yield as a result of biotic and abiotic stresses.

The more common methods employed for the identification of DNA markers are restriction fragment length polymorphism (RFLP), random amplified polymorphic DNA (RAPD) and amplified fragment length polymorphism (AFLP).

RAPD is a fast, easy and efficient method to obtain information on the genetic variation of the plants. It involves the use of "arbitrary" primers (which could be purchased from commercial sources) in a polymerase chain reaction (PCR), resulting in the amplification of several discrete DNA products. Using molecular genetic techniques, the responses of plants to specific abiotic stresses are now better understood. RAPD analysis has been used to determine the interspecific and intraspecific variations in different plant species (Autunes *et al.*, 1997 and Chandra Shekara *et al.*, 2005). RAPD analysis was used to screen for salt tolerance in maize callus lines (Zacchini *et al.*, 1997) and tomato (Foolad and Chen, 1998). The present investigation was aimed to study the genetic distances among different rice genotypes with response to salinity stress.

## MATERIALS AND METHODS

Seeds of rice (*Oryza sativa* L.) varieties *viz.*, Dandi, IR-36 and GR-3 were obtained from Main Rice Research Station, Navagam, Gujarat and CSR-1 from the Central Soil Salinity Research Institute, Karnal, Haryana. Seeds were incubated at $45^0$C for 48 hrs. in incubator to break the dormancy, soaked in distilled water for overnight and germinated in Petridishes. After sprouting seedlings were inserted and raised on nylon net kept over a Petridish filled with Yoshida nutrient solution. Seedlings were raised at two different salinity levels *i.e.* 3 and 5 EC and compared with control. DNA was extracted from fifteen days old seedlings.

*DNA Extraction :*

The genomic DNA was extracted from the seedlings of four rice varieties CSR-1, Dandi, IR-36 and GR-3, grown at 3 and 5 EC salinity in Yoshida nutrient solution with control (without salt) by a modified Cetyl Trimethyl Ammonium Bromide (CTAB) method (Keim *et al.*, 1988) with some modifications. Seedlings (500 mg) from each cultivar were powdered in liquid nitrogen using a pestle and mortar. The resulting powder was transferred to 10 ml centrifuge tube and extracted for 1 hr at 65°C with 5 ml of pre-warmed (65°C for 20 min) extraction buffer. Proteins were extracted with one volume of chloroform: isoamyl alcohol (24:1). The tubes were shaken by slowly inverting slants for 20 minutes. The tubes were centrifuged at 10,000 rpm for 10 min at 4°C and the aqueous layer was taken. Nucleic acids were precipitated with 0.7 volume of isopropanol, washed with 70% ethanol,

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