

## Molecular characterization among strains of chickpea root nodule bacteria isolated from different areas of middle Gujarat

V.R. HINGE\*, R.L. CHAVHAN<sup>1</sup>, Y.A. DESHMUKH<sup>2</sup> AND S.N. SALUNKHE<sup>3</sup>

Department of Agricultural Botany, B.A. College of Agriculture, Anand Agricultural University,  
ANAND (GUJARAT) INDIA

### ABSTRACT

Eighteen strains of Root nodule bacteria were collected from the chickpea plant, grown in different areas of middle Gujarat, viz., Anand, Dahod, Thasara, Arnej and Dhanduka. These strains were confirmed as *Rhizobium* by using different biochemical test and maintained their pure culture. Molecular characterization based on repetitive DNA sequence especially, ERIC sequence (Enterobacterial Repetitive Intergenic Consensus) were done together with two known *Rhizobium* strains, one commercial culture (GSFC, Vadodara), five standard strains of *Rhizobium* and one standard strain of *Agrobacterium tumefaciens*. The total of 320 no of amplicons was generated by using ERIC primer pair. The strain MTCC 4188 (*Mesorhizobium ciceri*) produced highest no of amplicons while strain MTCC 120 (*Bradyrhizobium japonicum*) showed a less no of amplicons. Data analysis of ERIC fingerprinting pattern clustered all RNB strains and standard strains into four major clusters as per their phylogenetic relationship. Majority of RNB strains (65 per cent) were closely related to the genus *Mesorhizobium ciceri* species and *Mesorhizobium loti*, while remaining 40 per cent RNB strains showed similarity to *Rhizobium leguminosarum* (MTCC 99) and *Agrobacterium tumefaciens* (MTCC 431). The ERIC-PCR fingerprinting could become a powerful tool for depicting the genetic diversity among eighteen RNB strains and standard strains. The data based on ERIC fingerprinting pattern could help to determine phylogenetic relationships among these RNB strains and will be helpful for development of diagnostic primer for identification of efficient strains of Chickpea Root Nodulating Bacteria.

**Key words :** Chickpea, *Rhizobium*, Root nodule bacteria, Phylogenetic

### INTRODUCTION

Chickpea (*Cicer arietinum* L.), belongs to the family *Leguminosae*, sub family *Papiliondiae*, tribe *cicerae*, is third most widely grown, self pollinated grain legume in the world. India is a premier chickpea growing country accounting for 67 per cent of total area and production of the world (Ali *et al.*, 2005). In Gujarat, the area of cultivation under chickpea is 170.0 thousand hectares, producing 140.0 thousand tonnes with productivity of 850 kg ha<sup>-1</sup> (Source <http://agricoop.nic.in/Agristatistics.htm>).

Besides wide nutritional and agriculture importance, Chickpea also plays an important role in improving soil fertility through the process of Biological Nitrogen Fixation (BNF). *Rhizobium* spp. mainly from the genus *Mesorhizobium* form nodules on chickpea root and fix atmospheric nitrogen symbiotically. Rainfed chickpea growing areas fall under Bhal and Coastal Agro climatic zone (zone VIII), where the soils are poor in drainage and saline, sodic or saline-sodic in nature and alkaline in reaction. The legume *Rhizobium* Symbiosis is affected by salinity, as it affects the growth of *Rhizobium*, nutrient uptake of plant and nodule formation mechanism. The yield and productivity of chickpea in these areas is affected due to environmental conditions such as drought and salinity. The chickpea variety GG-2 is mainly grown

in Bhal area. There is a tremendous scope to increase yield and production of chickpea in this area through improved efficiency of biological nitrogen fixation, which in turn will also reduce the requirement of nitrogenous fertilizer and ultimately cost the of production.

Diverse bacterial population is observed in root nodules, such as *Pseudomonas*, *Rhodopseudomonas*, *Agromyces*, *Bacillus*, *Microbacterium*, *Phyllobacterium* etc. (Zakhia *et al.*, 2006). Chickpea root nodules may contain some other endophytic bacteria, which help nodulation and nitrogen fixation. These bacteria (mainly *Bacillus* and *Pseudomonas*) promote nodulation by native *Rhizobia* and co-inoculation of such bacteria with effective *Rhizobium* strains of chickpea increase nodulation and nitrogen fixation significantly (Parmar *et al.*, 1999). These-helping bacteria may also promote the plant growth by solubilization of minerals such as phosphorus, production of siderophores, which solublize and sequester iron or production of plant growth regulators (Hormones) (Tilak *et al.*, 2005). The regions of middle Gujarat may contain strains of *Rhizobium*, well adapted to varying soil and environmental conditions. The *Rhizobium* strains isolated from these areas will be very effective for evaluating as a chickpea inoculant in saline areas of middle Gujarat, due to possessing capacity to survive or persist

\* Author for correspondence. <sup>1</sup>Department of Plant Biotechnology, College of Agricultural Biotechnology, LATUR (M.S.) INDIA

<sup>2</sup>MAHYCO, Life Sciences Research Centre, Crop Gene Function and Manipulation Lab, JALNA (M.S.) INDIA

<sup>3</sup>Department of Agricultural Botany, Marathwada Agricultural University, PARBHANI (M.S.) INDIA