ORIGINAL ARTICLE

Symbiotic potential, competitiveness and compatibility of indigenous *Bradyrhizobium japonicum* isolates to three soybean genotypes of two distinct agro-climatic regions of Rajasthan, India

M.K. Meghvansi *, Kamal Prasad, S.K. Mahna

Department of Botany, Maharshi Dayanand Saraswati University, Ajmer 305009, Rajasthan, India

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Symbiotic effectiveness; *Bradyrhizobium japonicum*; Inoculation; Soybean cultivars; Agro-climatic regions

**Abstract** In the current study, we recovered sixteen bradyrhizobial isolates from root nodules of two soybean genotypes (JS 335 and PK 472) grown in two distinct agro-climatic conditions (Bundi and Udaipur) of Rajasthan, India. Symbiotic effectiveness of these isolates was evaluated under greenhouse conditions. On the basis of statistical analysis of data (ANOVA followed by LSD \( P \leq 0.05 \)), four effective isolates namely BJ335-1, BPK-3, BPK-5 and UJ335-1 were screened out from the greenhouse experiment. The compatibility to three soybean genotypes, and the competitive ability with other field population of rhizobia, of these four isolates was further determined by conducting field trial. Results demonstrated significant variation in the symbiotic potential of tested isolates with respect to different soybean genotypes. Response of soybean genotype JS 335 towards inoculation was relatively better suggesting its suitability in the Haroti region of Rajasthan. Moreover, BJ 335-1 and BPK-3 isolates were found to be highly efficient as they significantly improved the nodulation, plant growth and seed yield. Possible factors responsible for variable response of bradyrhizobial isolates towards inoculation in three soybean genotypes are discussed. Further,
1. Introduction

Soybean (*Glycine max* (L.) Merr.) is the most important grain legume crop in the world in terms of total production and international trade. According to a report (Anon., 2007), world soybean production in year 2007–2008 was 220.81 Million Metric Ton (MMT). Soybean contains 40–42% protein and 18–22% oil comprising up of 85% unsaturated fatty acid and is free from cholesterol. Soybean protein provides all eight amino acids in the amount needed for human health, hence it is called meal of the field (Rathore, 2000). It is therefore, highly desirable in human diet and animal (Aslam et al., 1995; Haq et al., 2002). Soybean has a unique importance in the Indian agricultural economy because there is a great shortage of edible oil in the country. Since past few years, the consumption of oil has been increasing steadily as a result of rise in population and living standard of people, the basic question before the scientists is to develop a concrete strategy that permits self-reliance in edible oils (Mehgsvansi, 2006). India ranks fifth in soybean production in the world. The main soybean producing states of India are Madhya Pradesh, Maharashtra and Rajasthan while some soybean areas also exist in the states of Uttar Pradesh, Andhra Pradesh, Punjab, Tamil Nadu, Uttarakhand (now Uttarakhand), Gujarat, Karnataka and Chhattisgarh. Madhya Pradesh is the largest soybean producer state and is known as SOYA STATE of India (Mahna, 2005). In Rajasthan, soybean is mainly cultivated in the south eastern part of the state covering Kota, Bundi, Bharatpur and Jhalawar districts which is known as Haroti region (Mehgsvansi et al., 2005) while it is grown in patches in some other districts like Sawai Madhopur, Bhilwara, Chittorgarh, Dausa, Dungarpur, Banswara and Udaipur (SOPA 2000–2001; www.sopa.org as on 04.01.2006). Total soybean production in Rajasthan for the year 2007–2008 was 0.735 MMT in comparison to the national figure 9.473 MMT (www.sopa.org as on 15.04.2009).

The symbiosis between rhizobia and legume are a cheaper and usually more effective agronomic practice for ensuring an adequate supply of N for legume based crop (Zahran, 1999) and thus can play a significant role in improving the fertility and productivity of soils. Identification and selection of effective rhizobial strains are important for preserving them for future research. Since, the host-rhizobia relationship is more complicated (Lohrke et al., 1995) and is affected by several factors, the variation in the Inoculants performance is often encountered. Differences between *Bradyrhizobium* strains regarding their effectiveness with different soybean genotypes have been reported by several workers (Okereke et al., 2001; Tien et al., 2002; Mahna et al., 2006).

Better N₂ fixation can be achieved by selecting superior rhizobia. However, selection of these rhizobia would need to take into consideration not only their N₂-fixing capacity, but also competitive ability against native rhizobia which are frequently ineffective in N₂ fixation. Superior N₂-fixing strains have to outcompete native rhizobia and occupy a significant proportion of the nodules. For this to be achieved, rhizobia have to be selected under natural conditions in competition with the native rhizobia (Rengel, 2002). The subject of symbiotic effectiveness and competitiveness of rhizobia in Indian context assumes more significance and has attracted a lot of Indian workers (Shivananda et al., 2000; Appunu and Dhar 2006; Appunu et al., 2008). However, in Rajasthan, most of the rhizobial research has been confined to the tree legumes (Srivastava and Prabhakaran, 1999; Mahobia and Mahna, 2002; Mahobia, 2003) while little attention has been paid to the studies on rhizobia of soybean despite being an important oil yielding crop. As a consequence, symbiotic potential of the rhizobia autochthonous to different soybean growing regions of Rajasthan is still unexploited. In the context of these views, in the current investigation, collection of soybean root nodules from various cropping fields of Rajasthan was carried out, and bradyrhizobia were isolated and authenticated. The main objectives of the present study were (1) to evaluate symbiotic effectiveness of bradyrhizobial isolates by performing greenhouse experiment and (2) to determine their competitive ability with other field population of rhizobia and the compatibility to different soybean genotypes, under field conditions.

2. Materials and methods

2.1. Field sampling and isolation of bradyrhizobia

An extensive survey of soybean growing fields of Bundi and Udaipur districts of Rajasthan (see Fig. 1 for location map) was made during Kharif, 2003 at the 50% flowering stage of the crop and, soybean plants belonging to two important cultivars namely JS 335 and PK 472 were excavated at various field sites {Adaptive Trial Centre (ATC), Bundi and Rajasthan College of Agriculture (RCA), Udaipur} and were transported to the laboratory in plastic bags. Serial dilution agar plate method (Somasegaran and Hoben, 1994) was employed with 20E medium (Werner et al., 1975) for the purpose of isolation of bradyrhizobia from soybean root nodules. Isolates were streak purified on the same medium. All the isolates were subjected to their morphological, cultural and biochemical characterization as described in the literature (Vincent, 1970; Creager et al., 1990; Cappuccino and Sherman, 1992). Further, the isolates showing resemblance with *Bradyrhizobium japonicum* with regard to morphological, cultural and biochemical properties were subjected to authentication test in growth pouches (Mega International, USA) under controlled environmental conditions (day length, 14 h; temperature, 28 ± 1 °C, light intensity, 12,000 lux; humidity, 70–80%). Isolates were nomenclatured indicating their region of origin, name of cultivar and isolate number. Authenticated isolates were maintained as frozen glycerol stocks at −40 °C.

2.2. Symbiotic effectiveness of bradyrhizobial isolates

2.2.1. Greenhouse experiment (GHE)

Air-dried and sieved autoclaved soil (pH, 8.1; EC, 0.56 dS m⁻¹; OC, 0.17%; available N, 62.91 mg kg⁻¹ and Olsen-P,
16.00 mg kg\(^{-1}\) collected from non-legume cultivated land of Ajmer (Rajasthan) was used to fill in earthen pots (8 kg pot\(^{-1}\)). Seeds of two soybean genotypes (JS 335 and PK 472) were surface sterilized and then were inoculated with mid log phase culture of sixteen authenticated bradyrhizobial isolates using slurry method (Vincent, 1970). Amongst sixteen isolates, each strain was tested in its parent cultivar and uninoculated seeds of the same served as controls. Five seeds per pot were sown for each isolate and after emergence plants were thinned to two plants per pot. Pots were maintained in a greenhouse (light intensity 15,000–19,000 lux, temperature 27–35 °C and humidity 70–80%) and irrigated with sterile distilled water as needed. Experiment was performed in randomized complete block design with six replicates. After 45 days, plants were harvested and data related to nodulation and shoot biomass were collected. In addition, nitrogen content of dried plant shoots was estimated using macro-Kjeldahl procedure (Jackson, 1973).

2.2.2. Acetylene reduction activity

Seeds of two soybean genotypes (JS335 and PK 472) were surface sterilized with 4% NaOCl for 10 min and grown on water agar (1%) plates. 5–7 days old seedlings (with radicle length of 3–4 cm) were dipped into the exponentially growing broth culture of four bradyrhizobial isolates (BJ335-1, BPK-3, BPK-5 and UJ335-1). These isolates were not only tested in the soybean cultivar from which they were originally isolated but also tested heterologously in both the soybean genotypes (JS 335 and PK 472). The bacterized seedlings were then planted in the germination pouches and these pouches were placed in growth room for thirty days under the controlled environmental conditions as mentioned earlier for authentication test. Thirty days old, four root nodule systems (RNS) from each treatment were then exposed to the mixture of acetylene-in-air in assay vials (60 ml). These vials were sealed (subha seal) and 10% (6 ml) of air from these vials was replaced with equal volume of acetylene. Assay vials were then incubated for 30 °C. Ethylene production due to nitrogenase reduction, was assayed using gas chromatograph with a flame-ionization detector (FID) having a Porapak column. After incubation, 1 ml of gas from assay vials was injected into gas chromatograph with the help of syringe. Similarly values were recorded for standard ethylene gas (Hardy et al., 1968). Dry weight of oven dried (80 °C for 3 days) RNS was taken and the amount of C2H4 produced was calculated using the equation given below:

\[
\text{n moles of ethylene produced h}^{-1} \text{g}^{-1} \text{dry weight of RNS} = \frac{V \times R}{T \times DW},
\]

where, \(V\) = volume of air space in assay vial; \(R\) = chromatograph reading; \(T\) = time of incubation in h; \(DW\) = dry weight of RNS (mg).

The temperature of injection port and FID detector was 110 °C and the column temperature was 80 °C. The flow rate

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**Figure 1** Location map. (1) Sampling site Bundi; (2) sampling site Udaipur; (3) research lab Ajmer.
50 ml min\(^{-1}\) of carrier gas nitrogen was used. Air and hydrogen gas were adjusted to flow at the rates of 300 and 90 ml min\(^{-1}\), respectively.

2.2.3. Field trial

Experiment was performed in 29.00 \(\times\) 11.00 meter area with the layout randomized complete block design with treatments in a split plot arrangements using three soybean cultivars (JS 335, JS 71-05 and NRC 12) and four bradyrhizobial isolates (BJ335-1, BPK-3, BPK-5 and UJ335-1), at ATC Bundi during Kharif, 2004. Each isolate was tested in three soybean cultivars individually with its respective control. For one isolate, 480 seeds were sown in four rows and row length was kept 3 m with 35 cm distance between two adjacent rows. Data on nodule and shoot biomass were collected after 45 days of the trial while seed yield was recorded at the crop maturation stage.

2.3. Statistical analysis of data

Standard errors of means were calculated when appropriate and analysis of variance was carried out for the data generated from greenhouse experiment and field trial, and means were separated by the least significant difference (LSD) test (Sokal and Rohlf, 1981).

3. Results

3.1. Field sampling and Isolation of bradyrhizobia

From the root nodules of soybean plants of two distinct regions, a total of nineteen bacterial isolates were obtained amongst which eight isolates of JS 335 cultivar from Udaipur region, two isolates of JS 335 cultivar and nine isolates of PK 472 cultivar from Bundi region were recovered. Not much variation in the colony shape and texture of the studied bacterial isolates was recorded. These colonies did not absorb congo red dye present in the medium. Seventeen bradyrhizobial isolates out of nineteen, demonstrated alkaline reaction in Hofer’s alkaline medium (HAM) with slight variability in terms of intensity of colour, while two isolates exhibited acidic reaction. All the tested isolates were able to reduce the nitrate present in the medium with slight strain variability. Though in general, production of H\(_2\)S gas was negative for all the isolates but a very slight variability could also be observed where three isolates gave 95% negative results while remaining isolates showed 100% negative test. Methyl red test was negative for all the isolates but again a slight strain variability was noticed for these two tests as in methyl red test one isolate showed 90% negative, two showed 95% negative and the remaining showed 100% negative test. Variability was noticed in citrate utilization test also where two isolates showed 90% negative, one showed 95% negative and the remaining showed 100% negative test (Table 1). In plant assay test, sixteen isolates were authenticated as *Bradyrhizobium* which were taken into account for further investigation in detail. Three isolates (including two acid producing isolates) showing negative result were discarded.

3.2. Symbiotic effectiveness of bradyrhizobial strains

3.2.1. Greenhouse experiment

In the GHE successful nodulation was observed in all the soybean genotypes raised after inoculation with bradyrhizobial isolates and as expected no nodules were found on the uninoculated plant roots. Amongst all the tested sixteen isolates, maximum nodule biomass was recorded in the JS335 genotype inoculated with BJ335-1 strain. Nodular biomass was found to be relatively higher in genotype JS 335 compared to cultivar PK 472. In addition, majority of the bradyrhizobial isolates significantly improved the shoot biomass of the plants of two soybean genotypes. The most significant enhancement in shoot biomass over control was recorded due to inoculation with BJ335-1 strain. Nodular biomass was found to be relatively higher in genotype JS 335 compared to cultivar PK 472. In addition, majority of the bradyrhizobial isolates significantly improved the shoot biomass of the plants of two soybean genotypes. The most significant enhancement in shoot biomass over control was recorded due to inoculation with BJ335-1 strain. Nodular biomass was found to be relatively higher in genotype JS 335 compared to cultivar PK 472. In addition, majority of the bradyrhizobial isolates significantly improved the shoot biomass of the plants of two soybean genotypes.

| Brady-rhizobial strains | Parent cultivar | Region of origin | Growth on HAM | Nitrate reduction | H\(_2\)S production | Methyl red test | Citrate utilization |
|-------------------------|----------------|------------------|---------------|------------------|-------------------|-----------------|--------------------|
| BJ 335-1                | JS 335         | Bundi            | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| BJ 335-2                | JS 335         | Bundi            | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| BPK-1                   | PK 472         | Bundi            | + + + +        | + + + +          | -ve*              | -ve*            | -ve                |
| BPK-2                   | PK 472         | Bundi            | + + + +        | + + + +          | -ve               | -ve             | -ve**              |
| BPK-3                   | PK 472         | Bundi            | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| BPK-4                   | PK 472         | Bundi            | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| BPK-5                   | PK 472         | Bundi            | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| BPK-6                   | PK 472         | Bundi            | + + + +        | + + + +          | -ve**             | -ve**           | -ve                |
| BPK-7                   | PK 472         | Bundi            | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| BPK-8                   | PK 472         | Bundi            | + + + +        | + + + +          | -ve*              | -ve*            | -ve                |
| UJ 335-1                | JS 335         | Udaipur          | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| UJ 335-2                | JS 335         | Udaipur          | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| UJ 335-3                | JS 335         | Udaipur          | + + + +        | + + + +          | -ve**             | -ve**           | -ve                |
| UJ 335-4                | JS 335         | Udaipur          | + + + +        | + + + +          | -ve               | -ve             | -ve                |
| UJ 335-5                | JS 335         | Udaipur          | + + + +        | + + + +          | -ve**             | -ve**           | -ve                |
| UJ 335-6                | JS 335         | Udaipur          | + + + +        | + + + +          | -ve               | -ve             | -ve                |

\(-ve\) Negative test; \(-ve*\) 95% negative test; \(-ve**\): 90% negative test.

'+ + + +' 100% Positive test; '+ + +' 95% positive test; 'HAM' Hofer’s Alkaline medium.
Symbiotic potential, competitiveness and compatibility of indigenous *Bradyrhizobium japonicum* isolates

307

Figure 2  Effect of bradyrhizobial inoculation on nodulation and growth of soybean genotype JS 335 under greenhouse conditions. Values without common letters differ significantly at LSD *P* ≤ 0.05.

![Graph showing nodulation and growth of soybean genotype JS 335](image)

Figure 3  Effect of bradyrhizobial inoculation on nodulation and growth of soybean genotype PK 472 under greenhouse conditions. Values without common letters differ significantly at LSD *P* ≤ 0.05.

![Graph showing nodulation and growth of soybean genotype PK 472](image)

3.2.2. Acetylene reduction activity (ARA)

ARA for root nodule systems (RNS) infected with four bradyrhizobial isolates ranged from 8.49–11.77 nmol C\textsubscript{2}H\textsubscript{4} h\textsuperscript{-1} mg\textsuperscript{-1} rns dw and 7.84–12.21 nmol C\textsubscript{2}H\textsubscript{4} h\textsuperscript{-1} mg\textsuperscript{-1} rns dw for JS 335 and PK 472 genotypes, respectively. Furthermore, the values of ARA recorded for the RNS infected with BJ335-1 and BPK-3 isolates were significantly high compared to those recorded for the RNS treated with the remaining isolates (Fig. 4).

Figure 4  Acetylene reductase activity (ARA) of root nodules systems (rns) of two soybean genotypes inoculated with four bradyrhizobial isolates under controlled conditions. Values without common letters differ significantly at LSD *P* ≤ 0.05 (A & E- BJ335-1; B & F-BPK-3; C & G- BPK-5; D & HUJ335-1). Values for A–D recorded in cv. JS 335 and value for E–H recorded in cv. PK 472.

![Graph showing ARA activity](image)

4. Discussion

The morphological, cultural and biochemical properties of the bacterial isolates were quite similar to those recorded for *Bradyrhizobium* as described earlier by several other workers (Jordan, 1984; Werner, 1992; Srivastava et al., 1995). Mahna et al. (2003) also reported that cells of bradyrhizobial strains were Gram negative, long, motile rods and formed small circular, glistening, whitish colonies on 20E medium which is parallel to the results obtained in the current study. Similarly, Somasegaran and Hoben (1994) suggested that typical rhizobial colonies should show little or no congo red absorption in dark. In the current study, seventeen isolates showed alkaline reaction in BTB test (growth on HAM) which is supported by the results of Hameed et al. (2004) who also reported similar observations for rhizobial strains of soybean recovered from arable fields of Pakistan. As per the information available on [http://www.bio.net/bionet/mm/microbio/1997-December/011013.html](http://www.bio.net/bionet/mm/microbio/1997-December/011013.html) accessed on 20.04.08 soybean *Bradyrhizobium* gives positive catalase, oxidase and nitrate reduction tests, which strengthens the present findings. In addition, indole production for all the bradyrhizobial isolates was positive.
in the present investigation, which is in accordance with the earlier findings of Hameed et al. (2004). Statistical analysis (ANOVA followed by LSD P ≤ 0.05) of the data collected from the greenhouse experiment clearly showed that bradyrhizobial inoculation with two soybean genotypes (JS 335 and PK 472) improved the nodulation, plant biomass and shoot N. These submissions aligned correctly with previous findings of several other workers (Egamberdiyeva et al., 2004; Schulz et al., 2005; Cassán et al., 2009). Although no direct nitrogen fixation data were collected in this experiment, higher nitrogen accumulation in shoots of plants raised after bradyrhizobial inoculation compared to control suggests greater nitrogen fixation. Lukwati and Simanungkalit (2002) also found the significant increase in the dry matter, nitrogen and phosphorus uptake of soybean plants by bradyrhizobial inoculation in sterilized soil. In the current study, high nodulation was observed in the greenhouse experiment which might be attributed to the lack of antagonism in the autoclaved soil used in the greenhouse experiment. Similar to this result, Zdor and Pueppke (1988) also reported more nodules in autoclaved soil while studying the interaction of soybean and B. japonicum 123 and 138 serogroups. A continuous and coordinated selection of the most effective combinations of host and microbial symbionts is a prerequisite for profitable and sustainable agricultural systems (Rengel, 2002). The performance of tested isolates was quite variable and soybean genotype JS 335 responded in a better way in comparison to cultivar PK 472 towards inoculation. Milic et al. (2002) while studying the activity of nitrogen fixation and nitrogen assimilation enzymes in soybean plants inoculated with B. japonicum strains, also reported variability in the performance of different bradyrhizobial strains in terms of dry matter mass and nitrogen content in the nodules of the soybean varieties. Such variations may be attributed to the variations in the genomic constitutions of the host or bacteria or both which control symbiosis or there might be more than one affinity group within the legume rhizobia leading to such variation. In the present investigation, considerable cultivar effect on the nodulation and seed yield caused by various bradyrhizobial isolates was noticed. These results are consistent with those of Okereke et al. (2001) who reported that inoculation response of Bradyrhizobium in different soybean cultivars was cultivar and site-specific. According to Rengel (2002), there is genotypic variation in the germplasm of legume species in all components of the signaling pathway. There is generally a gene(s)-for-gene interaction between rhizobia and the host. The genotypic specificity of the nodulation process depends on such an interaction. On the contrary, Lukwati and Simanungkalit (2002) reported insignificant variation in interactions between varieties of soybean and bradyrhizobial inoculation. This suggests the need for careful Bradyrhizobium strain selection for improving inoculation response in soybean cultivars. The fact that inoculation response was cultivar specific suggests that strategies for improving inoculation response in soybean cultivars should also consider the soil environment where the soybean is to be produced. Comparative assessment of results of the greenhouse experiment and field trial suggested that there was consistency in the performance of two bradyrhizobial isolates BJ335-1 and BPK-3 which resulted into significant improvement in nodulation and field trial suggested that there was consistency in the performance of two bradyrhizobial isolates BJ335-1 and BPK-3 which resulted into significant improvement in nodulation, vegetative growth and seed yield of three soybean genotypes. Increased nodulation and subsequent nitrogen fixation resulted in the measured increases in plant growth and grain yield. Similar to the present results, Egamberdiyeva et al. (2004) noticed positive effect of inoculation of B. japonicum S2492 on growth, nodule number and yield of soybean under field conditions. Furthermore, they reported that yield of soybean varieties was 48% higher for inoculated plant than the uninoculated ones. Likewise, Simanungkalit et al. (1998) conducted a field experiment to evaluate the effectiveness of twentyfour bradyrhizobial strains and reported that ten strains amongst them increased soybean yield by 38%. Comparable to this, in the current study inoculation of two efficient isolates namely BJ335-1 and BPK-3 in three soybean genotypes brought about significant enhancement in the seed weight ranging from 18.03% - 42.13% over control while performance of remaining two strains was not satisfactory. Oker-
eke et al. (2001) in a field experiment also reported that when soybean cultivars were inoculated with foreign bradyrhizobia in two locations in south east of Nigeria, variation in strain performance was noticed which was also evident in the current study. In the present study, non-indigenous isolate (UJ335-1) could not compete with the indigenous strains, which is a very important criterion for the selection of efficient strains and weeding out of less effective ones. Somewhat parallel to it, Osunde et al. (2003) found the foreign strains appeared to be less competitive, but more effective, than the indigenous populations. Low level of competitive ability of non-indigenous isolate might be attributed to its limited gene pool for adapting to local conditions as opposed to indigenous strains which are well adjusted to prevailing conditions at the site and, moreover, can adapt to just about any change in conditions (Vlassak and Vanderleyden, 1997). While conducting field trial no fertilizer was applied to the field so that the test strains could derive their nitrogen requirement through BNF and exhibit their maximum potential by establishing effective symbiosis with soybean host and ultimately improve the soil fertility. Also, it is well established that addition of N fertilizer can lead to lower production of nodules in legumes (Rubio Aris et al., 1999; Vargas et al., 2000). While comparing the overall performance of four bradyrhizobial isolates in three soybean genotypes under field conditions, two isolates namely BJ335-1 and BPK-3 were identified as superior strains in terms of their symbiotic effectiveness, competitiveness and their compatibility with three soybean genotypes. Amongst three soybean genotypes tested under field conditions, JS335 showed better interaction with the bradyrhizobial isolates, which was evident from the relatively higher values of the studied parameters. Moreover, under the INCO-DEV Research project two consecutive field trials were conducted (data not given here) at private farm, Marwara Chauki, Kota (Rajasthan) during Kharif, 2004 and Kharif, 2005, and the results showed the consistency of BJ335-1 in terms of its efficiency. Also, these trials confirm the better adaptability of cultivar JS 335 under edapho-climatic conditions prevailing in Kota region. In field trial 2004, performance of bradyrhizobial strain BJ335-1 was superior in its cultivar group of five strains and inoculation of this strain caused 35.38%, 17.38%, 32.27% and 21.73% enhancement in nodule biomass, plant biomass, shoot N and seed weight of cultivar JS 335, respectively, compared to control. Moreover, BJ335-1 exerted highly significant influence in enhancing the studied parameters of soybean crop in field trial conducted at ATC, Bundi during Kharif, 2005. This also exhibits the suitability of BJ335-1 to be used as bioinoculant for soybean in Kota region as well as Bundi region (Mahna, 2006).

5. Conclusions

In general, the inoculation of soybean seeds with population of reasonably effective and persistent bradyrhizobial strains improved the soybean yield. Based on the evidences generated from various experiments performed in the present investigation, following conclusions/recommendations are made:

There exists a considerable variability amongst the autochthonous bradyrhizobial isolates of different genotypes of soybean grown in two distinct agro-climatic conditions (Bundi and Udaipur) of Rajasthan with respect to their symbiotic interaction. Looking to the response of soybean genotype JS 335 towards inoculation in greenhouse experiments and field trials, it is recommended that farmers should be encouraged to use this cultivar to enhance the area of soybean cultivation as well as its production in Rajasthan.

Effective bradyrhizobial isolates BJ 335-1 and BPK-3 may be utilized for inoculants production at large scale and the multilocational trials are required to determine their suitability for other agro-climatic conditions.

Consistency in performance of bradyrhizobial BJ335-1 isolate in Bundi as well as Kota field trial also clearly indicates its adaptability in both the regions.

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