CHAPTER V

DISCUSSION

The results on isolation of *Rhizobium* and field studies on evolution of different isolates' performance are discussed below:

Background of the study:

The present investigation was an important part of study, in order to select effective rhizobial isolate(s) for enhancing productivity of neglected but very important leguminous crop of Chhattisgarh like Horsegram (Kulthi) grown in different seasons under different climatic conditions of Chhattisgarh state. Initially, the area of Chhattisgarh which required the horse gram rhizobial inoculation was identified during isolation of *Rhizobium* germplasm from Sarguja, Korba and Bastar districts of Chhattisgarh. Thereafter, a series of studies under controlled and natural field conditions were planned to select effective rhizobial isolate(s) for Horsegram growers of this State. Present studies include conduction of preliminary screening experiments under controlled condition and screened out 223 Horsegram rhizobial isolates out of total 313 local isolates, on the basis of quality and quantity of nodulation.

Under the present investigation, during preliminary and first stage screening, 90 better isolates were identified out of total 313 newly obtained Horsegram *Rhizobium* isolates from the soils and plant samples of Sarguja, Korba and Bastar districts of Chhattisgarh.
Thereafter, a total of 90 isolates were further compared in glass house conditions during the second stage screening under the present investigation.

A. Study under controlled conditions:

Data presented in the Tables 2, 3, 4; Plate 3 and Appendix I & II indicated that per cent presence of native *Rhizobium* in soils of Sarguja, Korba and Bastar districts is much higher (80.26%) and only 19.74% area of these districts are devoid of native *Rhizobia* which must be needed for inoculation of *Rhizobium* for enhancing productivity of Horsegram crop (kulthi).

This observation is in close agreement with Tilak, 1991; Gupta *et al.*, 1995; Anonymous, 1996; Katre *et al.*, 1997 and Gupta and Jaggi, 1997 in case of Groundnut crop. Similar findings were also reported by Pimental *et al.*, 1990, who clearly concluded that water stress in soil, significantly reduced the soil rhizobial population, nodulation and ultimately the number of seeds per plant. Adverse effect of temperature on nodule formation and development have also been very well documented in clover: (Pankhurst and Gibson, 1973); pea (Frings, 1976); lotus and Stylosanthes (Rao, 1977) and soybean (Munever and Wollum, 1981; Badawy *et al.*, 1991).

This observation was also in line with that of Dudeja and Khurana, 1989; Gupta *et al.*, 1995 and Katre *et al.*, 1997. Gupta and Jaggi, 1997 who also clearly mentioned that soils of rainfed region like Chhattisgarh which is exposed to extreme dry and hot climate during summer, resulting into the loss of organic matter and population of mesophilic heterotrophs including *Rhizobium*.
In the second phase of study under controlled conditions which was carried out for screening of local \textit{Rhizobium} isolates, on the basis of degree of nodulation, dry weight of nodule biomass accumulation and nitrogen uptake at 60 days old Horsegram plant var. AK-31 (Batzli et al., 1992 and Kumar and Shrivastava, 1994).

**B. Glass House and Field Study:**

In soils like Inceptisol with low organic matter content having comparatively very low or ineffective naturally occurring population of \textit{Rhizobium}, response of introducing these new effective local \textit{Rhizobium} isolates could be expected. In order that such inoculations are successful, it is imperative that inoculated \textit{Rhizobium} isolates are well adapted to the soil and host and are able to compete with the indigenous soil microflora (Wilson, 1984). Both glass house and field experiments were conducted with 90 \textit{Rhizobium} isolates identified with potential to prove successful with Horsegram grown under climatic conditions of Chhattisgarh region. Glass house experiments have proved valuable for providing information with higher precision and lesser risk than field investigations, especially with reference to microbial inoculation studies without fear of contamination from inoculated experimental units during different agricultural operations etc.

Even though, a higher precision in results can be expected in glass house experiments over field investigations but effect of microbial inoculations in practical agriculture with competition with the indigenous soil microflora can be evaluated only through field experiments, conducted using standard recommended practices. Apart from above, it is also not easily possible to exploit crop beneficial microbes like \textit{Rhizobium} fully under glass house
conditions because of the restrictions imposed owing to smaller volume of sand /soil in glass house conditions available for development and extension of plant roots.

(I.) Glass House Study:

(a) Nodulation study:

(i) Number of nodules:

The data presented in table 5 (Plate 5 and Appendix - VI) indicated that number of nodules were obtained from 1.33 to 48.00 per plant due to inoculation of seeds with local isolates under the glass house study. It was observed that local isolate no 56 and 59 produced highest no. of nodules in the glass house grown 60 days old plant followed by isolate no. 41. Similar findings were also reported by Prasad and Ram (1986), Schroder (1989), Manoharn et al. (1990), Alagawadi et. al. (1993) and Devi and Gupta (1996). They mentioned number of nodules can be increased by inoculation with effective rhizobial isolates.

(ii) Weight of nodules:

The dry weight of nodules was obtained from .031 mg to 17.9 mg per plant (table 5 and Plate 5) due to inoculation of seeds with local isolates under the glass house study. It was observed that local isolate no. 59 produced the highest dry weight of nodules in the glass house grown 60 days old plant followed by isolate no. 29 and 47. This observation was in close agreement with Wankhade et al. (1992) who reported that number of root nodules and their weight per plant were significantly increased up to 75 DAS by rhizobial seed inoculation.
(b) **Plant height study:**

The data presented in table-6 (Plate 4 and Appendix- VI) revealed that height increased significantly from 15 to 30 DAS, 30 to 45 DAS and from 45 to 60 DAS due to inoculation of rhizobial isolates. However, plant height recorded maximum at 60 DAS due to inoculation of most of the rhizobial isolates tested under study. This may be because of greater rate of nutrient absorption up to reproductive phase i.e. up to 60 DAS (Thimme Gowda, 1993).

At 60 DAS, all the 90 isolates tested under the study gave higher nodulation plant growth possibly because of low and less effective native population of horse gram *Rhizobium* in the soil. Similar findings were also reported in by Siddaramaiah and Bagyaraj (1980), Prasad and Ram (1986), Schrodhu (1989), Manoharn et al. (1990), Alagawadi et al. (1993), Barik and Mukherjee (1995), Katre et al. (1997) and Prasad (1997). They clearly mentioned root nodulation including plant growth can be increased by inoculation with effective rhizobial isolates.

(c) **Bio-mass and N-uptake study:**

The performance of local isolate no. 59, 56 and 47 in terms of most effective parameters related to biological nitrogen fixation like dry weight of plant, nitrogen content and nitrogen uptake (Table-7 and Plate 5) was found higher among 90 isolates tested under study. The effectiveness of isolate no. 56, 59 and 47 under glass house conditions were also supported by data of nodulation study (table-5, Plate 5. and Appendix - VI). These findings were clearly supported by findings of Gupta et al. (1996).

Results of present investigation are also in confirmation with the findings of Siddaramaian and Bagyaraj (1981), Kumar and Srivastava (1994)
and Prasad (1997) who got better local isolates with those of recommended national and international isolates through systematic screening of local Rhizobium germplasm.

(II.) Field study:

It is known that a variety of factors can modify the effectiveness of rhizobial isolate(s) for a specific season and a specific region. The number of factors becomes more under field conditions to affect the performance or activity of rhizobial isolate(s). Therefore a field experiment was planned under different agro-climatic conditions for screening of rhizobial isolates under natural field conditions. (Batzli et al., 1992 and Katre et al., 1997).

(a) Plant height and nodulation study:

The data presented in table-8 (Fig.2) revealed that plant height increased significantly at 45 DAS due to inoculation of rhizobial isolates tested under study. This may be because of greater rate of nutrient absorption up to reproductive phase i.e. up to 45 DAS (Thimmegowda, 1993).

The data on nodulation at 45 DAS (Table-9, Fig. 3) further revealed that number of nodules increased significantly over control due to inoculation of 10 better local selected isolates studied under field conditions of different agro-climatic region.

Similarly, the dry weight of nodules also increased at 45 DAS due to inoculation of seeds with rhizobial isolates tested under study. This observation was in close agreement with Wankhade et al. (1992) who reported that number of root nodules and their weight per plant were significantly increased up to 75 DAS by rhizobial seed inoculation in groundnut.
At 45 DAS, all the 10 better isolates tested under the study gave significantly higher nodulation over control, possibly because of low and less effective native population of horsegram *Rhizobium* in the soil. Similar findings were also reported by Prasad and Ram (1986), Schrodu (1989), Manoharn *et al.* (1990), Alagawadi *et al.* (1993), Barik and Mukherjee (1995), Katre *et al.* (1997) and Prasad (1997). They clearly mentioned that root nodulation including plant growth can be increased by inoculation with effective rhizobial isolate.

(b) Biomass study (shoot weight):

The data presented in table-10, (Fig. 5) clearly showed that dry biomass of plant increased significantly at 45 DAS due to inoculation of ten better isolates, including application of General Recommended Dose. Similar findings were also reported by Nanja Reddy *et al.* (1992), Thimmegowda (1993), Ravi Kumar *et al.* (1994) and Mahakulkar *et al.* (1994). They mentioned that dry matter accumulation increased with the advancement of crop age. The accumulation increased rapidly up to 80 days after sowing followed by a decrease during the later stages of crop growth due to shedding of leaves.

Performance of isolates numbered 56 and 59 at Sarguja and Bastar in case of Raipur, isolate no.41, in terms of parameters like dry weight of plant at 45 DAS was found in better ten category of promising isolates tested under field conditions. The effectiveness of these isolates are also supported by data of nodulation study (Table 6 and 10). Results of the present investigations are in confirmation with the findings of Siddaramaiah and Bagyaraj (1981), Kumar & Shrivastava (1994) and Prasad (1997). They got better local isolates with those
of recommended national and international isolates through systematic screening of local rhizobial isolates.

(c) Yield study:

The data on seed and straw yield at harvest of crop (Table 16 and 17, Fig. 11 and Fig. 12) revealed that seed yield of Horsegram increased significantly due to use of ten better local isolates tested under study. These findings were similar as reported by Siddaramaiah and Bagyaraj (1980), Sridar et al. (1989), Tiwari et al. (1989), Wankhade et al. (1992), Balasubramanian and Palaniappan (1994), Patel and Thakur (1997) and Hegde (1998). They clearly concluded that inoculation with efficient Rhizobium isolates significantly increased the seed and straw yield of kharif Horse gram under field conditions.

Performance of local isolate no. 56 in terms of seed and straw yield was found superior among the ten better isolates studied. Similar views were also expressed by Siddaramaiah and Bagyaraj (1980) and Kumar and Srivastava (1994). Katre et al. (1997) also expressed the similar views and mentioned that local isolates are more effective for a particular agroclimatic region than the isolates imported from other places in general.

(d) N-uptake study:

Performance of local isolate No. 56 in terms of most effective parameters related to biological nitrogen fixation like per cent nitrogen content and its uptake at Harvest (Table-11 to Table – 14 and Fig 6 to Fig 9) was found the highest among all ten isolates including General Recommended Dose of fertilizer followed by isolate no. 11, may be due to their suitability at Sarguja, Bastar and Raipur districts concluded during the present investigation.
Findings of the present investigation also clearly showed that nitrogen content in Horsegram at harvest and N-accumulation was found higher in seed rather than Horsegram straw. At harvest stage of the crop growth, it was worthwhile to note that nitrogen content reached to its minimum in Horsegram straw. This is because of its high mobility towards seed from rest of the plant body. This observation is in close agreement with Thimmegowda (1993) and Patra et al. (1998). They clearly mentioned that nitrogen concentration in plant decreased steadily with the commencement of reproductive phase. Due to high mobility of nitrogen, it was translocated to the pod and concentration in seed increased. This showed that the pod nitrogen is derived from foliage. They further mentioned that the amount of nitrogen moved to the pod is proportionately lower to the quantity of nitrogen available in the foliage. It may be attributed that some quantity of nitrogen was being utilized in the increase of dry weight of vegetative part.

The performance of isolate no-56 in terms of effective BNF parameters like dry matter yield, seed and straw yield, nitrogen content in seed and straw and total nitrogen uptake by the crop at harvest, was found superior under natural field conditions among all ten selected isolates at all three locations, possibly because of its ability for competition with natural soil micro flora and its suitability for hot and humid climate of Chhattisgarh region.

From the present investigation, it was concluded that local isolate no. 56 was the most effective nitrogen fixer followed by isolate no-11 for Kharif Horsegram cultivations at all three location of different agro-climatic regions of Chhattisgarh.