RESPONSE OF BLACKGRAM (*Vigna mungo* L.) TO *BRADYRHIZOBIUM* INOCULATION AS INFLUENCED BY PHOSPHORUS LEVELS

By

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CERTIFICATE

This is to certify that thesis entitled "RESPONSE OF BLACKGRAM (*Vigna mungo* L.) TO *BRADYRHIZOBIUM* INOCULATION AS INFLUENCED BY PHOSPHORUS LEVELS" submitted to the Department of Soil Science, Sher-e-Bangla Agricultural University, Dhaka-1207 in partial fulfillment of the requirements for the degree of Master of Science (M.S) in Soil Science embodies the result of a piece of *bonafide* research work carried out by Mohammad Yusuf, Registration No. 00830 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that such help or source of information, as has been availed of during the course of this investigation has duly been acknowledged by him.

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Dedicated to

My

Beloved Parents and Nephew

Wasir Mohammad

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ABBREVIATIONS, ACRONYMS AND SYMBOLS

AEZ	Agro-Ecological Zone
ANOVA	Analysis of Variance
App.	Appendix
B	Boron
BARC	Bangladesh Agricultural Research Council
BARI	Bangladesh Agricultural Research Institute
BAU	Bangladesh Agricultural University
BAUR	Bangladesh Agricultural University *Rhizobium*
BBS	Bangladesh Bureau of Statistics
BCR	Benefit and Cost Ratio
BNF	Biological Nitrogen Fixation
BRRI	Bangladesh Rice Research Institute
C	Carbon
CD	Cowdung
CEC	Cation Exchange Capacity
cm	Centimeter
cmol kg^{-1}	Centimole per kilogram
Cu	Copper
CV	Coefficient of Variation
DAS	Days After Sowing
DM	Dry Matter
DMRT	Duncan's Multiple Range Test
DW	Dry Weight
EC	Electrical Conductivity
FAO	Food and Agriculture Organization
Fe	Iron
FYM	Farm Yard Manure
g	Gram
K	Potassium
kg	Kilogram
kg ha^{-1}	Kilogram per hectare
mg	Milligram
mm	Millimeter
Mn	Manganese
MP	Muriate of Potash
N	Nitrogen
nm	Nanometer

P	Phosphorus
ppm	Parts Per Million
PSO	Principal Scientific Officer
RCBD	Randomized Complete Block Design
S	Sulphur
SSO	Senior Scientific Officer
t	ton
t ha^{-1}	Ton per hectare
TSP	Triple Superphosphate
USDA	United States Department of Agriculture
Zn	Zinc
$\mu g\ g^{-1}$	Microgram per gram

ABSTRACT

A pot culture experiment was conducted at the Net House and Laboratory of Soil Science Division in Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur during September to December 2007 to evaluate the performance of phosphorus levels and *Bradyrhizobium* on growth, nodulation, yield, phosphorus, nitrogen content and uptake and other yield contributing characters of blackgram variety BARI Mash-1. *Bradyrhi zobium* inoculant (BARI RVm-301) was used for the above experiment. There were ten treatment combinations with two *Bradyrhizobium* namely non-inoculated and inoculated and five levels of phosphorus @ 0, 10, 20, 30 and 40 kg ha^{-1} with four replications. Significant influences of phosphorus level were observed in nodulation and yield (seed, pod and stover) and nutrient content and uptake by seed and other yield contributing characters. Application of phosphorus up to 20 kg ha^{-1} progressively increased the values of all the parameters. Further, higher doses application of phosphorus decreased the value of all of these parameters. *Bradyrhizobium* inoculant significantly increased nodule number and weight, root and shoot weights, root and shoot lengths, seed yield, pot yield, stover yield, plant height, 100-seed weight, pods $plant^{-1}$, seeds pod^{-1}, N, P content in seed and N, P uptake by seed, protein content in seed and protein yield compared to non-inoculation. *Bradyrhizobium* inoculation along with application of phosphorus @ 20 kg ha^{-1} produced the highest nodulation, yield and other parameters compared to other treatment combinations.

Chapter 1
Introduction

CHAPTER I

INTRODUCTION

Pulse, a protein rich agricultural crop, plays an important role in human nutrition. It is the cheapest source of protein for the poor people. Pulses, being leguminous crops, are capable of fixing atmospheric nitrogen in the soil and enrich soil fertility. Thus they are considered as soil fertility building crops (Kumar *et al.*, 1963). Some of the pulse seeds are consumed when they are at green stage. But it is taken mostly in the form of soup and "dal". Sometimes it is grown as green manuring crops and cover crops (Shaikh, 1977). The green plants, the dried stems and leaves after separation of grain and the husks of seeds are the valuable food to the livestock. It is an excellent source of easily digestible protein.

Annual crop legumes, grown in rotation with cereals crops, can increase yields of the cereals and contribute to the total N pool in the soil. Reported yield responses to previous legume crops are mainly in the range of 50-80% yield increase over control in cereal-cereal sequences (Evans *et al.*, 1991; Oikeh *et al.*, 1998). Benefits of legumes have been attributed to control of cereal diseases and insect pests, and improvements in soil structure (Reeves *et al.*, 1984).

Blackgram (*Vigna mungo* L.) is one of the major pulse crops grown in Bangladesh. It belongs to the family Legurminoceae and sub-family Papilionaceae. It is widely grown in the Indian subcontinent as a source of protein. These legumes can obtain nitrogen (N) from the atmosphere by fixation in their root nodules in symbiosis with soil rhizobia, and thus have the potential to yield well in N-deficient soils (Hafeez *et al.*, 1988). This characteristic is particularly important in developing countries due to the relatively high cost and/or restricted availability of chemical N fertilizers. Furthermore, incorporation of residues into the soil from a N-fixing legume crop may provide organic N for the subsequent benefit of a cereal crop (Rosales *et al.*, 1998).

Blackgram (*Vigna mungo* L.) is one of the widely grown pulse crops in Bangladesh for human consumption, animal fodder as well as soil fertility building purpose. But high cost and environmentally risky chemical fertilizers cause serious and continuous problem for increasing blackgram production in developing countries including Bangladesh. This problems are likely to become serious in future. Biological nitrogen fixation (BNF) resulting from symbiosis between legume crops and root nodule bacterium *Rhizobium/Bradyrhizobium* can ameliorate the situation by reducing the N-fertilizer inputs required to ensure productivity.

Blackgram is one of the most important pulse crops although not in terms of area (23,482 ha) and production (17, 000 t) but its consumption is quite high as a common pulse in Bangladesh (BBS, 2006). It is an important source of protein and several essential micronutrients. It is used as 'dhal' and as ingredient in preparation of many foods in terms in Bangladesh.

Phosphorus (P) is one of the essential mineral macronutrient, which is required for maximum yield of agriculturally important crops. Most of the essential plant nutrients, including phosphorus, remain mostly in insoluble form in soil (Abd-Alla, 1994; Yadav and Dadarwal, 1997). Ironically, soils may have large reserve of total phosphorus, but the amounts available to plants are usually a tiny proportion of thus total (Stevenson and Cole, 1999). Most agricultural soils contain large reserves of phosphorus, a considerable part of which has accumulated as a consequence of regular applications of P fertilizers (Richardson, 1994). However, a large portion of soluble inorganic phosphorus applied to soil as chemical fertilizer is rapidly immobilized or fixed as insoluble forms soon after application and becomes unavailable to plants (Dey, 1988; Singh and Kapoor, 1994; Yadav and Dadarwal, 1997). Farmers are thus asked to apply several-fold excess phosphorus fertilizers in order to overcome this problem. Therefore, the release of insoluble and fixed forms of phosphorus is an important aspect of increasing soil phosphorus availability.

Availability of P is a serious problem, because it is fixed in the soil and lowers the utilization efficiency of added P fertilizers by plants. In most tropical and sub-tropical soils, phosphate are predominantly present in the form of inorganic compounds, which belong to two groups those of calcium and those of iron and aluminum. Iron and aluminum phosphates predominate under acidic conditions. Plant takes up P in the form of soluble orthophosphate ions $H_2PO_4^{-1}$ and HPO_4^{-2}. The availability of these ions to plants depends mainly on soil pH (Nath and Borach, 1983). Soil microorganisms play a significant role in mobilizing P and increasing the availability for plants. The rhizobial association with cropping system has recently assumed important in the mineral nutrition of plants particularly with respect to phosphorus and other micronutrient.

Now a days, a number of organisms like *Rhizobium* has been identified to use as biological agent for fixing atmospheric nitrogen by process with legume crops and make available to the plants. Bangladesh Agricultural Research Institute (BARI) has isolated some *Rhizobium* strains for some pulse corps. It has already selected some *Bradyrhizobium* strains especially for blackgram varieties. To reduce the production cost and to fulfill the demand, more pulse production could be achieved through seed inoculation with *Bradyrhizobium* strains which is known to influence biological fixation, growth and yield of pulses. In Bangladesh, inoculation with *Bradyrhizobium* increased 77% dry matter production, 64% grain yield and 40% hay yield over non-inoculated control (Chanda *et al*., 1991). Maximum yields were obtained when fertilizers are applied along with *Rhizobium* inoculation (Bali *et al.*, 1991).

By growing blackgram in Bangladesh, there is a large scope of utilizing the biological nitrogen fixing technology for obtaining protein rich food legume and also to improve nitrogen fertility of the soil of this country.

Unfortunately, there is a lack of sufficient research work on the nutrient requirement particularly phosphorus and the effect of seed inoculation of blackgram with effective rhizobial strain for successful cultivation of this crop in Bangladesh. The present investigation was, therefore, undertaken to evaluate the response of blackgram to inoculation with *Bradyrhizobium* as influenced by phosphorus levels with the following objectives:

i) To investigate the effect of *Bradyrhizobium* inoculation on the growth, nodulation, yield, nitrogen and phosphorus uptake and other yield contributing characters of blackgram.

ii) To assess the response of blackgram regarding growth, nodulation, yield, nitrogen and phosphorus uptake to P application.

iii) To determine the relationship among different parameters of blackgram.

Chapter 2
Review of Literature

CHAPTER II

REVIEW OF LITERATURE

Biofertilizers are microorganisms which benefit the plants by increasing availibility of nitrogen or phosphorus or rapid mineralization of organic materials. Of the biofertilizes, the use of *Bradyrhizobium* was studied in Bangladesh to some extent. Only limited number of research works has so far been carried out on the combined use of *Bradyrhizobium* inoculation and P application on blackgram (*Vigna mungo* L.) varieties and other pulse crops. However, available information on the contribution of *Bradyrhizobium* inoculation and P application on blackgram has been reviewed in this chapter.

2.1 Effect of phosphorus on legumes

Effect of different levels of phosphorus on blackgram and other legumes have been presented below:

Mishra (1993) carried out a field experiment on sandy loam soil on farmer's field during the rainy seasons of 1986-87 at Sidhi, Madhya Pradesh where 3 blackgram (*Vigna mungo*) cultivars was given 0, 20, 40 and 60 kg P_2O_5 ha^{-1} and reported that 0, 20, 40 and 60 kg P_2O_5 ha^{-1} gave mean seed yields of 592, 655, 751 and 846 kg ha^{-1}, respectively.

Sharma *et al.* (1995) conducted a field trial in the monsoon season of 1991 at Gwalior, Madhya Pradesh with blackgram (*Vigna mungo*) cv. JU-2 treated with 0, 15 or 30 kg N, 0, 30 or 60 kg P_2O_5 and 0 or 60 kg S ha^{-1}. Application of N and P, either alone or with S, increased Mn, Zn, Cu and Fe contents in seeds and straw and the available Mn and Zn content in soil. Application of 30 kg N + 60 kg P_2O_5 + 60 kg S ha^{-1} gave the highest trace element content in blackgram. Soil available Cu content decreased with increasing N and P applications but increased with S application. Soil available Fe increased with increasing N and P applications and decreased with increasing S applications.

Bhalu *et al.* (1995) conducted a field experiment during the rainy season of 1990 at Junagadh, Gujarat with blackgram (*Vigna mungo*) and seed was inoculated with *Rhizobium* or not inoculated and given 10, 20 or 30 kg N and 20, 40 or 60 kg P_2O_5 ha^{-1}. Seed inoculation increased seed yield (471 vs. 434 kg ha^{-1}). Seed yield increased with up to 20 kg N (464 kg) and 40 kg P_2O_5 (475 kg). N and P uptakes and seed protein content increased with increasing N and P rates. Net return was the highest with seed inoculation.

Ali *et al.* (1995) carried out field trials at Mianchannu in 1992 and Layyah in 1993 on sandy loam soils low in OM, N and P and *V. mungo* was given no fertilizers or 50 kg N, 50 kg N + 50, 75, 100 or 125 kg P_2O_5 or 50 kg N + 125 kg P_2O_5 + 50 kg K_2O ha^{-1}. NPK gave the highest number of pods plant^{-1} (23.03-23.75) and seed yield (1080-1082 kg ha^{-1}) but was not significantly better than 50 kg N + 75 kg P_2O_5, which gave the highest 1000-seed weight (49.30 and 42.75 g in the 2 trials, respectively). Straw yields did not differ significantly among the treatments. Seed protein content was the highest with NPK.

In a field experiment during kharif 1995 at Shillongani, Assam, India, blackgram cv. T-9 was given 0, 20, 40 or 60 kg P ha^{-1} and 0, 25, 50, 75 or 100% of the lime requirement (4.04 t ha^{-1}) (Borah *et al.*, 2000). They observed that soil pH increased and all forms of soil acidity decreased with increasing lime rate, while exchangeable acidity and total acidity decreased with increasing P rate. The decrease in soil acidity was accompanied by reduction in the forms of Al with application of lime and P. However, lime x P interaction was not significant except for exchangeable acidity. Seed yield increased with increasing P and lime rates.

Trivedi (1996) carried out field trials in the rainy seasons of 1990-91 at Gwalior, Madhya Pradesh, India with *P. mungo* (*Vigna mungo*) cv. Jawahar Urd-2 and was given 0-30 kg N, 0-60 kg P_2O_5 and 0 or 60 kg S ha^{-1}. Seed yield, net returns and N, P and S contents in seed increased with rate of N, P and S applications.

Chaudhary and Das (1996) conducted an experiment in Uttar Pradesh, India and found that P, S and Mo application significantly increased the canopy, nodule count, yield of rainfed blackgram (*Vigna mungo*), yield of succeeding safflower and reduced splash loss and conserved more soil water. Water stable aggregates, infiltration rate, organic carbon, total N, available P, K, S and Mo in soil increased considerably after the harvest of blackgram but decreased after the harvest of succeeding safflower. Plant canopy showed significant positive relationship with nodule count, soil water conservation, water stable aggregates and infiltration rate but showed significant negative relationship with splash loss.

Rao and Rao (1996) performed a pot experiment with blackgram (*Vigna mungo*) and greengram (*V. radiata*) where soil was inoculated with the mycorrhizal fungi *Acaulospora spinosa*, *A. morrowae* or *Glomus epigaeum* (*G. versiforme*). P was applied as superphosphate, rock phosphate or tricalcium phosphate. In the inoculated treatments, root colonization was 68-89% in blackgram and 69-93% in greengram; colonization in both species was the lowest when pots inoculated with *A. morrowae* were given superphosphate, while the best colonization was given by *G. epigaeum* without added P. Shoot DW was increased by both mycorrhizas and applied P, and was highest with superphosphate + *G. epigaeum*. This treatment also gave the highest plant N and P concentrations and 100-seed weight.

Trivedi *et al.* (1997a) conducted a field experiment to study the effect of nitrogen, phosphorus and sulfur on yield and nutrient uptake of blackgram (*Vigna mungo*) at Gwalior, Madhya Pradesh during the 1990-91 kharif (monsoon) seasons. Application of 30 kg N, 60 kg P_2O_5 and 60 kg S ha^{-1} increased yield, net profit and nutrient uptake.

Trivedi *et al.* (1997b) carried out a field experiment on blackgram (*Vigna mungo*) where 0, 15 or 30 kg N ha^{-1}; 0, 30 or 60 kg P_2O_5 ha^{-1} and 0 or 60 kg S ha^{-1} were given on sandy loam soil at Gwalior, Madhya Pradesh, during the rainy seasons (kharif) of 1990 and 1991. Increasing levels of N, P_2O_5 and S significantly increased the seed and stover yields. Increases in mean seed yield over controls were 217, 273 and 109 kg ha^{-1} with 30 kg N, 60 kg P_2O_5 and 60 kg S ha^{-1}, respectively. The application of N with S significantly enhanced the yield. Increasing levels of N, P_2O_5 and S significantly increased the total uptake of N, P and S. The maximum net return (Rs. 3893 ha^{-1}) and BCR (2.01) was obtained with $N_{30}P_{60}S_{60}$.

Ramamoorthy *et al.* (1997) conducted a field trial at Pudukkottai in the 1992-93 rainy seasons and reported that yield of blackgram (*Vigna mungo*) increased up to 40 kg P_2O_5 ha^{-1} and the highest S rate of 40 kg ha^{-1}.

Mahmud *et al.* (1997) carried out an investigation during the rainy seasons of 1990 and 1991 on a silt loamy soil at Samaru, Zaria (11 degrees 11'N, 07 degrees 38'E, 686 metres above sea level) in the Northern Guinea savanna zone of Nigeria to study the response of blackgram (*Vigna mungo*) to three levels of phosphorus (0, 13 and 26 kg P ha^{-1}) and three intra-row spacing (10, 20 and 30 cm). Plant height, weight of seed $plant^{-1}$, weight of pods $plant^{-1}$, 1000-seed weight and seed yield were significantly increased with increasing phosphorus application from 0 to 26 kg P ha^{-1}. Increasing intra-row spacing increased weight of seed $plant^{-1}$, weight of pods $plant^{-1}$ and 1000-seed weight but decreased plant height and seed yield per unit area.

Tomar (1998) performed a field experiment in Madhya Pradesh, India, during the rainy season of 1994-95 and 1995-96 to evaluate the effect of phosphate solubilizing-bacteria and farmyard manure on the yield of blackgram (*Vigna mungo*) under different fertility levels. Yield and yield components of cv. RU 2 increased significantly with the application of

N, P and K at 20, 26.20 and 16.66 kg ha^{-1}, respectively. Phosphate-solubilizing bacteria inoculation at 10 g kg^{-1} seed and farmyard manure at 5 t ha^{-1} both singly and in combination gave a significant increase in yield and its attributes compared with the control. Application of phosphate-solubilizing bacteria combined with farmyard manure gave the grain and straw yields of 819 and 1200 kg ha^{-1}, respectively. Application of N, P, and K at 20, 26.20 and 16.66 kg ha^{-1}, respectively, phosphate-solubilizing bacteria inoculation with farmyard manure gave the highest grain yield (1001 kg ha^{-1}).

Gunjkar *et al.* (1999) carried out an experiment on blackgram (*Vigna mungo*) at Parbhani during kharif (monsoon) 1994 and reported that blackgram gave seed yields of 658, 870 and 921 kg ha^{-1} with 0, 25 and 50 kg N ha^{-1}, and 768, 800, 836 and 863 kg ha^{-1} with 0, 25, 50 and 75 kg P_2O_5 ha^{-1}, respectively.

Thakur (1999) conducted field experiments at Tendani, Chhindwara (Madhya Pradesh) during the rainy seasons of 1991 and 1992 to evaluate the effects of P, S and *Rhizobium* on growth and yield of blackgram. Inoculation of *Rhizobium* culture on the surface of dry seeds before sowing helped to improve the seed and straw yields. Significant increases in seed and straw yields were observed up to 40 kg P_2O_5 and 20 kg S ha^{-1}, mainly due to improvement in plant height, branches $plant^{-1}$ and pods $plant^{-1}$.

Thiyageshwari and Perumal (2000) conducted a pot experiment with a Vertic Ustropept to test the changes in soil phosphorus forms, uptake and grain yield due to integrated nutrient management of blackgram through conjunctive use of imported Tunisia rock phosphate, vermicompost and phosphobacteria. Growth of blackgram and P uptake was slow in the vegetative phase but rapid during the reproductive phase. Vermicompost application significantly enhanced grain yield followed by phosphobacteria above 100% P as Tunisia rock phosphate. Phosphorus uptake by blackgram was higher in the combined application of rock phosphate with vermicompost and phosphobacteria. Available phosphorus was higher in the vegetative stage and later decreased at harvest due to P utilization by blackgram.

Shrivastava and Rajput (2000) conducted field experiments during the kharif seasons of 1995 and 1996 at 11 and 13 locations in Madhya Pradesh, India to study the response of phosphate solubilizing bacteria (PSB) on the seed yield of blackgram. PSB + recommended fertilizer treatment recorded the highest yield (1109 kg ha^{-1}) and maximum net profit (Rs 8148 ha^{-1}), which was on a par with the PSB + 3/4 recommended fertilizer treatment, but superior to the other treatments.

Kumar *et al.* (2000) conducted a field experiment with urdbean (*V. mungo*) cv. Pant U-19 on a sandy loam soil, in Ranchi, Bihar, India, during 1996. The treatments consisted of 4 phosphate rates (0, 20, 40 and 60 kg P_2O_5 ha^{-1}), applied at the time of sowing. The greatest plant height (63.60 cm), number of branches $plant^{-1}$ (5.98), number of pods $plant^{-1}$ (35.90), number of seeds pod^{-1} (5.60), 1000-grain weight (37.90 g), number of leaves $plant^{-1}$ (18.92), leaf area $plant^{-1}$ (139.80 dm^2) and grain yield (14.60 q ha^{-1}) were observed at 40 kg P_2O_5 ha^{-1}, which was significantly superior to the rest of the treatments. At 20 kg P_2O_5 ha^{-1}, these parameters were significantly superior to the control.

Maqsood *et al.* (2001) carried out a study in a sandy clay loam field to investigate the effect of phosphorus rates on the agronomic traits of two mashbean (*V. mungo*) genotypes (Mash-97 and Mash-88) at the University of Agriculture, Faisalabad, Pakistan, during 1998. The phosphorus rates were 0, 50, 75 and 100 kg P_2O_5 ha^{-1}. Mashbean genotypes did not differ significantly regarding number of plants m^{-2}, plant height, number of seeds pod^{-1}, total number of seeds $plant^{-1}$, 1000-seed weight, seed yield and harvest index. However, Mash-97 gave significantly more seeds $plant^{-1}$ than that of the Mash-88. Phosphorus application at 75 kg ha^{-1} gave significantly the highest seed yield of 1832 kg ha^{-1} against the minimum of 1390 kg ha^{-1} without phosphorus.

Tomar *et al.* (2001) conducted a field experiment at the G.B. Pant University Research Station, Ujhani, Uttar Pradesh, India, during kharif 1994-95 to study the effect of *Rhizobium*, vesicular arbuscular mycorrhiza (VAM, *Glomus caledonium*) and phosphate solubilizing bacteria (PSB, *Pseudomonas striata* strain P-27) inoculation, with and without P, on blackgram (*Vigna mungo*) seed yield. Phosphorus application in soil with medium P content (5.4 mg kg^{-1}) increased nodulation, grain yield, N and P in plant and grain over no phosphorus control. Forty kilograms of P_2O_5 per hectare recorded an increase of 20.6% in nodule dry weight, significant increases of 0.35 g kg^{-1} in N concentration and 1.28 g kg^{-1} in P concentration of plant over 20 kg P_2O_5 ha^{-1}. Similar significant increases of 0.59 g kg^{-1} in grain yield and 0.54 and 0.23 g kg^{-1} in N and P concentrations of the grain, respectively, over 20 kg P_2O_5 ha^{-1} were also obtained with higher dose. Inoculation of *Rhizobium* + VAM + PSB at all the stages of plant growth recorded maximum increases in all the parameters studied. *Rhizobium* gave the highest and 21.0% more nodule number, 34.7% more nodule dry mass, 0.73 g kg^{-1} more N in grain and 4.2% higher grain yield over PSB.

Bhattacharyya and Pal (2001) performed a field experiment in West Bengal, India, during the pre-kharif season to study the effect *Bradyrhizobium* inoculation, P (at 0, 20 kg ha^{-1}) and Mo (at 0, 0.5 and 1 kg ha^{-1}) on the number of nodules $plant^{-1}$ of summer greengram cv. T-44. Inoculation and application of P and Mo significantly influenced the number of nodules per plant and plant height.

An investigation was carried out by Singh *et al.* (2002) to study the effect of N, P and K application on seed yield and nutrient uptake by blackgram at Central Agricultural University, Imphal, Manipur, India during 1998 and 1999. In the grain yield, response of blackgram to the various treatments combinations of N (0 and 15 kg ha^{-1}), P (0, 30 and 60 kg ha^{-1}) and K (0 and 20 kg ha^{-1}), the highest yield was obtained from the application of 15:60:20 kg $N:P_2O_5:K_2O$ ha^{-1} which was at par with control and this might be due to higher values of organic carbon, N, P_2O_5 and K_2O in the soil. The total uptake of nutrients by blackgram was associated with higher biomass production.

Srinivas and Shaik (2002) conducted a field experiment to find out the effects of N (0, 20, 40 and 60 kg ha^{-1}) and P (0, 25, 50 and 75 kg ha^{-1}) along with seed inoculation with *Rhizobium* culture on the growth and yield components of greengram. Plant height generally increased with increasing rates of P and with increasing rates of N up to 40 kg ha^{-1} followed by decrease with further increase in N.

Patel and Thakur (2003) performed a field experiment with blackgram, comprising of 4 P levels (0, 30, 60 and 90 kg ha^{-1}) with and without phosphate solubilizing bacteria (PSB; 5 kg ha^{-1}) and farmyard manure (FYM; 1 t ha^{-1}) during the rainy season of 1997 and 1998 in Raigarh, Madhya Pradesh, India. The soil was sandy loam in texture, slightly acidic in reaction, with low available N and P and medium available K. Yield attributes and yield were significantly affected by P, PSB and FYM applications. Application of 60 kg P ha^{-1} significantly increased the number of pods $plant^{-1}$, 100-seed weight and seed yield of blackgram over 30 kg P ha^{-1} and the control, but found at par with 30 kg P ha^{-1} for seeds pod^{-1} during both the years. Application of 60 kg P ha^{-1} recorded 10.4 and 69.3% higher yield over 30 kg P ha^{-1} and the control, respectively. Application of PSB and FYM resulted in significantly the highest pod length, pods $plant^{-1}$ and seeds pod^{-1} during 1997, and appreciably increased 100-seed weight and seed yield during both years. An increasing trend in harvest index was observed with increasing levels of P and application of PSB and FYM.

Sulphur-phosphorus interaction has been studied in field experiments on a soil deficient in S and medium in P with blackgram (*Phaseolus mungo*) as the test crop by Singh and Singh (2004). The treatments were taken in factorial combination of three levels of S (0, 30, and 60 kg ha^{-1}) and four levels of P (0, 30, 60 and 90 kg ha^{-1}) applied through gypsum ($CaSO_4.2H_2O$) and triple superphosphate, respectively. A uniform dose of N was applied with the treatments. The grain, straw and total dry matter yield increased with the application of S and P individually, but decreased when S and P were applied in different combinations. Sulphur application increased S and P content in seed as well as in straw. Total P content increased with applied P and decreased with S application. Applied S increased and P decreased the protein content in grains. Changes in N: S ratio in grain was affected by S and P application. The antagonistic effect of S and P fertilizers on uptake and utilization of each other was more conspicuous when both were applied together.

Poonkodi (2004) conducted a field experiment during 2000-01 in Annamalai Nagar, Tamil Nadu, India to investigate the effect of P and pressmud on nutrient uptake of blackgram and on the postharvest soil nutrient status. The treatments were: T_1: control; T_2: recommended P rate (RDP) at 100% as single superphosphate (SSP); T_3: RDP at 100% as diammonium phosphate (DAP); T_4: press mud at 6.25 t/ha; T_5: RDP at 100% as SSP + PM; T_6: RDP at 100% as DAP + PM; T_7: RDP at 75% as SSP + PM; T_8: RDP at 75% as DAP + PM; T_9: RDP at 50% as SSP + PM; and T_{10}: RDP at 50% as DAP + PM. T_5 produced the highest available N (167.7 kg ha^{-1}), while the lowest (155.9 kg ha^{-1}) was obtained under the control. The available P and K values showed similar patterns with that of the available N. The application of P as SSP or DAP at 75% + PM showed similar efficacy as RDP at 100% as DAP or SSP + PM in increasing the crop nutrient uptake and postharvest N status.

In a field experiment on sulfur (S) deficient soils (sandy loam) in India, the effects of S (0, 30 and 60 kg ha^{-1}) and phosphorus (P) (0, 30, 60, and 90 kg ha^{-1}) were studied for yield, nutrient uptake and quality of blackgram (*Vigna mungo*) by Singh (2004). Application of S up to 30 kg ha^{-1} enhanced the average grain yield of blackgram by 22.2% over control. The application of P up to 60 kg ha^{-1} increased the grain yield by 31% over control. On fitting the average grain yield of two years (1993-94 and 1994-95) data in to quadratic equation, the optimum dose of S for blackgram was found to be 34.6 kg ha^{-1}. Similarly, the optimum dose of P was found to be 64.5 kg ha^{-1} giving maximum grain yield of 10.27 q ha^{-1}. Similar trend in yield response of straw was also recorded. Total S uptake progressively increased from 5.37 to 6.62 with the increasing S levels. The uptake of P also increased with the levels of P. The uptake of P and S increased up to 30 kg and 60 kg ha^{-1} level, respectively. Protein content significantly increased with the increased doses of S and P over control.

2.2 Effect of *Bradyrhizobium* inoculation on legumes

Effect of *Brayrhizobium* inoculation on blackgram and other legumes have been presented below:

Satter and Ahmed (1995) carried out a field experiment on mungbean (*Vigna raidata* L.) to study the response of inoculation with *Bradyrhizobium* inoculants incorporating BINA 403, BINA 407, RCR 3824 and RCR 3825 strains as single and mixed culture. They observed that *Bradyrhizobium* inoculation increased the number of nodules and weight significantly compared to uninoculated treatments.

Thakur and Panwar (1995) conducted a field trial where *Vigna radiata* cv. Pusa-105 and PS-16 were given seed inoculation of both. They found that inoculation either singly in combination increased plant height compared with no inoculation.

Shukla and Dixit (1996b) conducted a field experiment where green grain cv. Pusa Baisakhi was seed inoculated with *Rhizobium* sown in rows, 20, 30 and 40 cm apart and given 0-60 kg P_2O_5 ha^{-1}. They found that seed inoculation increased seed yield.

Sharma and Khurana (1997) studied the effectiveness of single and multistrain inoculants in field experiment with summer mungbean variety SML-32 and found that grain yield was superior in multistrain inoculants. On an average, single strain and multistrain *Rhizobium* inoculants increased the grain yield by 10.4% and 19.3% over uninoculated control, respectively.

Patra and Bhattacharyya (1997) carried out a field trial with *Vigna radiata* cv. B-1, *Bradyrhizobium* and urea (25 kg ha^{-1}). They found that all the treatments increased nodulation compared to control and the highest nodules were found with *Bradyrhizobium* + urea.

Das *et al.* (1997) conducted field trials where *Vigna radiata* cv. Noyagrah local seed were inoculated with *Rhizobium* and/or VAM culture, which was applied at the rate of 15 kg ha^{-1}. They observed that number of nodules was increased with dual inoculation compared with uninoculated control.

Jayakumar *et al.* (1997) carried out a pot culture experiment where blackgram (*Vigna mungo*) plants were raised from: uninoculated seeds; uninoculated seeds with 250 g pot^{-1} coir-pith compost applied at 15 and 45 days after sowing; *Rhizobium*-inoculated, lime-pretreated seeds; and *Rhizobium*-inoculated seeds + lime-pretreatment with 250 g pot^{-1} coir-pith compost. Application of coir-pith compost increased root length more than shoot length of plants compared to controls. It is suggested that coir-pith compost improved the growth of blackgram by increasing the rate and activity of nodulation and increasing the availability of

P and K. *Rhizobium* inoculation with lime seed pelleting increased both root and shoot lengths by 45%. *Rhizobium* inoculation, lime pretreatment and coir-pith compost synergistically increased the dry weight of plants and number of nodules plant^{-1} compared to controls. Total nitrate content, nodule leghaemoglobin content and leaf nitrate reductase activity were higher in plants raised from *Rhizobium*-inoculated seeds, particularly with lime-pretreatment and coir-pith compost. It was concluded that pelleting seeds with lime ensures the survival of *Rhizobium*, which improves plant growth under stress.

Ghosh and Poi (1998) carried out a pot experiment where soybeans, groundnuts, mashkalai (*Vigna mungo*), mung (*Vigna radiata*) and lentil (*Lens esculentus*) were inoculated with *Bradyrhizobium*, *Bacillus polymixa* and *Glomus fasciculatum* in different combinations. They found that nodulation and population of microorganisms in the rhizosphere were highest from combined inoculation with all three microorganisms.

Maldal and Ray (1999) conducted a field experiment where mungbean cv. B105, B1 and Hoogly local were untreated, seed inoculated with *Bradyrhizobium* and 20, 30, or 40 kg N ha^{-1} as urea were given. The results revealed that nodulation was greatest with inoculation in B105 and Hoogly local while it was decreased by inoculation and N treatment in B105.

Sharma *et al.* (1999) performed a field experiment in 1997/98 in Himachal Pradesh and *V. mungo* cv. Pant U-19 was seed inoculated with one of eleven *Rhizobium* strains or not inoculated and given 0 or 20 kg N ha^{-1}. Seed yield was highest with inoculation with a local strain (1.30 t ha^{-1}). The application of 20 kg N gave higher seed yield than no N (1.24 vs. 1.14 t). Application of nitrogen and inoculation increased nodulation and nodule dry weight plant^{-1}, crop growth rate and relative growth rate. No significant effects were observed in protein content of seed and straw but a slight improvement was observed over control.

Kavathiya and Pandey (2000) conducted a pot experiment with *Bradyrhizobium* on seed inoculation of mungbean and found that nodule $plant^{-1}$ increased significantly over uninoculated control.

Choudhury *et al.* (2000) performed a field experiment during kharif season of 1999 with mungbean and seed inoculation with *Bradyrhizobium* strains. They found that dry matter production was increased by about 50% due to *Bradyrhizobium* inoculation.

Srivastav and Poi (2000) conducted field experiments to determine the symbiotic efficiencies of greengram (*V. radiata*) and blackgram (*V. mungo*) after inoculation with a native Bradyrhizobium strain and the residual effects of 7 *Bradyrhizobium* strains (NG-13/1, M-10, Kuthi AR-1, Jca-1, Caj-3, NK-4 and Caj6/1) in neutral pH soil, in Mohanpur, West Bengal, India. Symbiotic variations of greengram and blackgram were observed due to the host and inoculant strains. Inoculation with M-10 strain in greengram resulted in the highest dry matter production and nitrogen fixation, while NK-4 inoculation into blackgram resulted in the highest nitrogen uptake and grain yield. The residual potentialities of the 7 strains were very low in subsequent seasons; however, strains M-10 and NK-4 were better than the other strains. This was due to their higher adaptive nature and competitiveness over the native strains.

Sharma *et al.* (2000) carried out a field experiment during kharif 1997 at Palampur, Himachal Pradesh, India where *V. mungo* was seed inoculated with 1 of 9 *Rhizobium* strains and given 0 or 20 kg N ha^{-1}. Growth, yield and dry matter accumulation increased with N application and *Rhizobium* inoculation, with the local strain giving the best results.

A field experiment was conducted in Vamban, Tamil Nadu, India by Nagarajan and Balachandar (2001) during the kharif season of 1998 to study the effects of organic amendments on the nodulation and yield of blackgram cv. Vamban 1. The treatments consisted of *Rhizobium* (strains CRU 7 for blackgram and CRM 11 for greengram) seed inoculation, 15 t farmyard manure (FYM ha^{-1}, FYM + *Rhizobium*, 5 t compost ha^{-1} (prepared from leaves and twigs of *Sesbania sesban*, *S. grandiflora*, *Cassia fistula*, *Cass ia auriculiformis*, and *Claricidia* (*Gliricidia*) along with cowdung and rock phosphate), compost + *Rhizobium*, 5 t biodigested slurry ha^{-1}, and biodigested slurry + *Rhizobium*. In general, seed inoculation of *Rhizobium* and application of organic amendments enhanced biomass, root nodulation, and grain yield. Biodigested slurry at 5 t ha^{-1} + *Rhizobium* gave the greatest plant height (42.7 and 53.7 cm for blackgram and greengram, respectively), nodule number (23.3 and 24.0), nodule weight (45.3 and 42.3 mg), and grain yield (758.3 and 732.0 kg ha^{-1}).

Bhattacharyya and Pal (2001) conducted a field experiment in West Bengal, India, during the pre-kharif season to study the effect *Bradyrhizobium* inoculation, P (at 0, 20 kg ha^{-1}) and Mo (at 0, 0.5 and 1 kg ha^{-1}) on the number of nodules $plant^{-1}$ of summer greengram cv. T-44. Inoculation and application of P and Mo significantly influenced the number of nodules per plant and plant height.

Sharma (2001) carried out a field experiment in Palampur, Himachal Pradesh, India during the kharif season. Seeds of mungbean cv. Pusa Baisakhi were inoculated with three strains of *Bradyrhizobium* culture (Ludhiana, local isolated and IARI). The various physiological and yield parameters of mungbean were evaluated. Crop growth rate, relative growth rate, days to 50% flowering, days to maturity was at maximum when mungbean seeds were treated with the local isolate.

Tomar *et al*. (2001) conducted a field experiment at the G.B. Pant University Research Station, Ujhani, Uttar Pradesh, India, during kharif 1994-95 to study the effect of *Rhizobium*, vesicular arbuscular mycorrhiza (VAM, *Glomus caledonium*) and phosphate solubilizing bacteria (PSB, *Pseudomonas striata* strain P-27) inoculation, with and without P, on blackgram (*Vigna mungo*) seed yield. Phosphorus application in soil with medium P content (5.4 mg kg^{-1}) increased nodulation, grain yield, N and P in plant and grain over no phosphorus control. Forty kilograms of P_2O_5 each hectare recorded an increase of 20.6% in nodule dry weight, significant increases of 0.35 g kg^{-1} in N concentration and 1.28 g kg^{-1} in P concentration of plant over 20 kg P_2O_5 ha^{-1}. Similar significant increases of 0.59 g kg^{-1} in grain yield and 0.54 and 0.23 g kg^{-1} in N and P concentrations of the grain, respectively, over 20 kg P_2O_5 ha^{-1} were also obtained with higher dose. Inoculation of *Rhizobium* + VAM + PSB at all the stages of plant growth recorded maximum increases in all the parameters studied. *Rhizobium* gave the highest and 21.0% more nodule number, 34.7% more nodule dry mass, 0.73 g kg^{-1} more N in grain and 4.2% higher grain yield over PSB.

Singha and Sarma (2001) conducted an experiment in India on blackgram cv. T-9 to study the effect of different levels of P fertilization and *Rhizobium* inoculation of seeds on yield and nutrient uptake. Application of P significantly increased the grain and straw yield and N, P and K uptake. P at 45 kg ha^{-1} produced the highest grain and straw yield and was at par with the application of 25 and 35 kg P ha^{-1}. N uptake increased from 20 to 30 kg ha^{-1} with application of 25 to 45 kg P ha^{-1}, respectively. *Rhizobium* inoculation significantly increased the number (2.2%) and mass (9.5%) of root nodules $plant^{-1}$ compared to the control indicating increased efficiency of the crop to fix the atmospheric N.

The interaction effects of *P. fluorescens* with *Rhizobium* in the management of blackgram (*Vigna mungo*) root rot caused by *M. phaseolina* was studied under in vitro and in vivo conditions (Shanmugam *et al*., 2001). In a related experiment, a fluorescent pseudomonad strain (Pf 1) which effectively inhibited the mycelial growth of *M. phaseolina* was studied for its compatibility with *Rhizobium*. Dual cultures and calorimetric studies established the compatibility of *P. fluorescens* and *Rhizobium* in controlling the root rot pathogen. However, glass house and field studies showed that seed treatment and soil application of Pf 1 was the most effective treatment to reduce root rot incidence to increase yield.

The nodulation characteristics of 8 varieties of blackgram (*Vigna mungo*) were studied by Reddy and Mallaiah (2001). *Rhizobium* sp. was isolated from the T-9 cultivar of the crop. The effect of three different methods of *Rhizobium* inoculation on the nodulation of blackgram was studied. The initiation of nodulation was early and the numbers of nodules formed were more in the seed inoculation method than in soil inoculation or seedling inoculation methods. Three isolates of *Rhizobium*, viz. VM isolate, AH isolate and SG isolates, isolated respectively from blackgram, *Arachis hypogaea* and *Sesbania grandiflora*, were used to study their effect on nodulation and nitrogen content of blackgram cultivar T-9. In plants inoculated with the VM isolate, nodules appeared 12 days after sowing, and a maximum of 84 nodules plant^{-1} were found during the reproductive stage of the crop. The nitrogen content of the nodules at the reproductive stage was 4.5%. The nitrogen content of the shoot was 1.9% at the vegetative stage (25-day-old plants), 3.0% at the reproductive stage (45-day-old plants) and 1.0% at the harvesting stage. The nitrogen content of the fresh seeds was 5.78% in the inoculated plants, while that in uninoculated controls was only 2.72%. The plants inoculated with the AH isolate showed better nodulation and nitrogen content compared to the plants inoculated with the VM isolate. However, the SG isolate completely failed to produce nodules on blackgram.

Sarkar *et al*. (2002) inoculated the seed of blackgram with strains of *Bradyrhizobium* viz. M-10, 129-USA, 480-M, and MK-5 before sowing in a field experiment conducted to determine the suitable cultivars and *Bradyrhizobium* strain for use in the locality. Cultivars

M-16 recorded longer roots and higher root volume plant^{-1}, number of nodules plant^{-1} and test weight compared to A-43. The interaction effects between cultivar A-43 and *Bradyrhizobium* strain MK-5 resulted in the highest root volume plant^{-1} (1.297), number of nodules plant^{-1} (7.030) and test weight (4.227 g), whereas the interaction effects between cultivar A-43 and bradyrhizobial strain 480-M resulted in the longest roots (14.72 cm). Correlation coefficient studies showed high correlation between seed yield and high correlation between correction with test weight, shiot length and shoot fresh weight. Root length and root volume were inversely correlated with test weight.

Malik *et al.* (2002) studied the effects of seed inoculation with *Rhizobium* and P application (at 0, 30, 50, 90 and 110 kg ha^{-1}) on the growth, seed yield and quality of mungbean cv. NM-98 in a field experiment conducted at Faisalabad in Pakistan during the autumn of 2000. Plant height at harvest was the highest when inoculated with *Bradyrhizobium* (68.13 cm). Seed inoculation with *Rhizobium* and application of 70 kg ha^{-1} resulted in the highest number of pods plant^{-1} (22.47), number of seed pod^{-1} (12.06), 1000-seed weight (42.27g) and seed yield (1158 kg ha^{-1}).

Srinivas and Shaik (2002) studied the effects of N (0, 20, 40 and 60 kg ha^{-1}) and P (0, 25, 50 and 75 kg ha^{-1}) along with seed inoculation with *Rhizobium* culture on the growth and yield components of greengram were determined in a field experiment conducted during the kharif seasons. Plant height generally increased with increasing rates of P and with increasing rates of N up to 40 kg ha^{-1} followed by decrease with further increase in N. Number of seeds per pods, 1000-seed weight, seed and haulm yields generally increased. Seed inoculation with *Rhizobium* resulted in higher values for the parameters measured relative to the control. The interactions effects between N and P were not significant for the number pods plant^{-1}, pod length and seed haulm yield.

Chatterjee and Bhattacharjee (2002) studied the effects of inoculation with *Bradyrhizobium* and phosphate soluble bacteria (PSB) on nodulation and grain yield of mungbean cv. B-1 in field trial conducted in West Bengal. Seeds of mungbean were inoculated with strains of *Rhizobium*, i.e JCa-1 and M-10 strains, at a population of 28.20 x 10^6 and 32.66 x 10^6 cells ml^{-1}, respectively, phosphate solubilizing bacteria containing *Bacillus polymyxa* and *Pseudomonus striata* at a population of 7 x 10^8 cells ml^{-1} at the time of the sowing. The plants inoculated with *Bradyrhizobium* strains and PSB showed increased rate of nodulation and N content. The percentage increased in seed yield over control was observed to be highly significant in plants inoculated with *Bradyrhizobium* strains and PSB.

Parveen *et al.* (2002) conducted a field experiment to observe rhizospheric microorganisms on growth and yield of greengram (*Phaseolus radiata*). The treatments were single, dual and combined inoculants of *Bradyrhizobium*, *Azotobacter chroococcum* and *Aspergillus*. The maximum root dry weight (0.37 g $plant^{-1}$) and seed yield (6.6 g $plant^{-1}$) were observed with single *Bradyrhizobium* sp.

Osunde *et al.* (2003) tested the response of two mungbean cultivars (TGX1456-2E) and TGX1660-19F) to *Bradyrhizobium* inoculation in a two year trials in the farmers' fields of Nigeria. Cultivar effect of plant of plant height and nodulation number was significant only in the first cropping season of the trial. Inoculation increased 40% seed yield in the first cropping season, while no such yield differences occurred in the second season. The proportion of nitrogen derived from nitrogen fixation ranged from 27% to 50% in the both cropping seasons and this was dependent on crop management on the farmers' field, rather than any cultivar or inoculation effect.

Asraf *et al.* (2003) conducted a field experiment to observe the effects of seed inoculation of a biofertilizer and NPK application on the performance mungbean cv. NM-98 at Faisalabad in Pakistan. The treatments consisted of the seed inoculation of *Rhizobium phaseoli* singly or in combination with 20:50:0, 40:50:0 or 50:50:50 NPK kg ha^{-1} (urea), P (single super phosphate) and K (potassium sulphate) were applied during sowing. The tallest plants (69.93 cm) were obtained with seed inoculation + 50:50:0 kg NPK ha^{-1}. Seed inoculation + 50:50:0 or 50:50:50 kg ha^{-1} resulted in the highest number of pods $plant^{-1}$ (28.97, 56.00, 63.90 and 32.56, respectively) and seed yield (1053, 1066, 1075 and 1072 kg ha^{-1}). Harvest index was highest with seed inoculation in combination with NPK and 40:50:0 (25.23), 50:50:0 (24.70) or 50:50:50 (27.5). Seed inoculation along with NPK at 30:50:0 kg/ha was optimum for the production of high seed yield by mungbean cv. NM-98.

Manivannan *et al*. (2003) carried out field experiments during 1999/2000 in Tamil Nadu, India to investigate the effect of foliar application of Microsol B (NPK with chelated micronutrients) with and without *Rhizobium* seed inoculation on the productivity of blackgram (cv. ADT3). The treatments consisted of foliar application of DAP at 30 and 45 days after sowing (DAS), Microsol B at 15, 30 and 45 DAS with and without *Rhizobium* seed treatment and an untreated control. *Rhizobium* seed treatment and foliar application of Microsol B recorded markedly higher leaf area index, dry matter production, crop yield, net assimilation rate, crop growth rate and relative growth rate in both years. No significant difference in relative growth rate was observed between treatments at the early stages, i.e. vegetative and flowering.

A study was conducted by Kumari and Nair (2003) to isolate efficient native strains of *Rhizobium* or *Bradyrhizobium* spp. and to develop suitable package of practices recommendations for their efficient use. The initial isolation of *Bradyrhizobium* spp. was done from seven different locations in Kerala, India, where the soil was generally acidic in nature. A total of 26 isolates (13 each from blackgram (*Vigna mungo*) and greengram (*V.*

radiata) were obtained and were screened for nodulation efficiency. The experiment was conducted in complete randomized block design with three replications for each isolate using unsterilized soil of pH 4.89 without any amendments such as applications of FYM or chemical fertilizers. It was also repeated under amended soil conditions using sterilized soil and application of FYM at 20 tonnes ha^{-1}. The selected isolates were further evaluated under field (Vellayani and Kayamkulam) conditions along with a package of practices recommendation (POP) developed by the Kerala Agricultural University. The extent of root nodulation, plant growth and yield were more in blackgram and greengram where *Bradyrhizobium* inoculation was done along with the POP recommendation. At Vellayani, the nodule number, plant dry weight and yield in blackgram were significantly high in the treatment combination of POP KA-F-B-6. At Kayamkulam, significant increases were obtained only in nodule number, nodule dry weight and yield. The results indicated that for acidic soils, the mere development of efficient native strains of *Rhizobium* or *Bradyrhizobium* alone was not sufficient but it should be along with a package of practices recommendation consisting of application of organic manure and liming to neutralize the soil pH.

Sriramachandrasekharan and Vaiyapuri (2003) coducted a pot culture experiment to study the effect of carbofuran in association with *Rhizobium* on the nodulation, growth, and yield of blackgram cv. ADT 3. Plant height (40.3 cm), number of nodules $plant^{-1}$ (36.4), effective number of nodules $plant^{-1}$ (24.5), nodule dry weight (20.2 g), root length (23.5 cm), shoot weight (0.82 g), root weight (0.32 g), number of pods pot^{-1} (18.2), pod yield pot^{-1} (55.6 g), root weight (0.32 g), and stover yield pot^{-1} (90.1 g) was the highest with 2.50 ppm carbofuran and decreased thereafter with further increase in carbofuran concentration. Irrespective of the levels of carbofuran, *Rhizobium*-inoculated blackgram showed better growth and higher pod yield (50.3 g) and stover yield pot^{-1} (81.1 g) than the uninoculated crop.

2.3. Interaction effect of phosphorus and *Bradyrhizobium* on legumes

Interaction effect phosphorus and *Brayrhizobium* inoculation on blackgram and other legumes have been presented below:

Mathan *et al.* (1996) conducted a field experiment during the monsoon seasons of 1991-92 at Coimbatore, Tamil Nadu where a total of 7 treatments compared the effect of applying in various combinations N, P, K, FYM, NAA and seed inoculation with Rhizobium were used on blackgram (*Vigna mungo*) cv. CO 5 yield. In 1992, seed treatment with fungicides was also included. The application of 25 kg N as urea + 50 kg P_2O_5 ha^{-1} as single superphosphate + 750 kg enriched FYM + 6.25 t FYM + foliar application of 25 kg diammonium phosphate at flower initiation and 15 days later + seed inoculation with *Rhizobium* produced the highest seed yields of 0.72 t ha^{-1} in 1991 and 0.62 t in 1992, compared with the control yields of 0.42 and 0.40 t, respectively. The seed crude protein content was increased by 14.5 and 15.4% in the highest yielding treatment compared with the controls.

Thakur (1999) conducted field experiments at Tendani, Chhindwara (Madhya Pradesh) during the rainy seasons of 1991 and 1992 to evaluate the effects of P, S and *Rhizobium* on growth and yield of blackgram. Inoculation of *Rhizobium* culture on the surface of dry seeds before sowing helped to improve the seed and straw yields. Significant increases in seed and straw yields were observed up to 40 kg P_2O_5 and 20 kg S ha^{-1}, mainly due to improvement in plant height, branches $plant^{-1}$ and pods $plant^{-1}$.

Twelve treatment combinations comprising 3 levels of phosphorus (0, 13.1 and 26.2 kg P/ha in the form of single superphosphate), 2 levels of farmyard manure (0 and 10 t/ha) and 2 levels of phosphate solubilizing bacteria (PSB; inoculated and uninoculated) were tested by Reddy and Swamy (2000) in blackgram (*Vigna mungo*) grown on a sandy loam at

Rajendranagar College, Hyderabad in summer 1999. With an increase in phosphorus level from 0 to 26.2 kg P ha^{-1}, yield attributes like pods $plant^{-1}$, seeds pod^{-1}, seed yield and haulm yield ha^{-1} increased by 53.5, 15.3, 50.6 and 26.3%, respectively, over the control. But seeds/pod was comparable with that of 13.1 kg P ha^{-1}. P content in haulm and seed increased significantly by the application of 26.2 kg P ha^{-1}. Farmyard manure at 10 t ha^{-1} increased dry matter $plant^{-1}$, pods $plant^{-1}$ and seed yield by 8.3, 9.2 and 6.5%, respectively, over no farmyard manure application. P content in haulm and seed increased due to farmyard manure. Soil inoculation with PSB gave increases in dry matter $plant^{-1}$, pods $plant^{-1}$ and seed yield of 6.6, 7.7 and 7.2%, respectively, over non-inoculation. Interaction of phosphorus with farmyard manure was significant with respect to seed yield. Economic analysis of the different treatments shows that the highest additional seed yield and additional net returns (Rs 3528 ha^{-1}) were associated with 26.2 kg P ha^{-1} + PSB inoculation + no farmyard manure, giving a benefit:cost ratio of 2.69.

Singha and Sarma (2001) conducted an experiment in India on blackgram cv. T-9 to study the effect of different levels of P fertilization and *Rhizobium* inoculation of seeds on yield and nutrient uptake. Application of P significantly increased the grain and straw yield and N, P and K uptake. P at 45 kg ha^{-1} produced the highest grain and straw yield and was at par with the application of 25 and 35 kg P ha^{-1}. N uptake increased from 20 to 30 kg ha^{-1} with application of 25 to 45 kg P ha^{-1}, respectively. *Rhizobium* inoculation significantly increased the number (2.2%) and mass (9.5%) of root nodules $plant^{-1}$ compared to the control indicating increased efficiency of the crop to fix the atmospheric N.

The effects of P (0, 20, 40 or 60 kg P_2O_5 ha^{-1}) and biofertilizers as seed inoculants (*Rhizobium* and the phosphate-solubilizing bacterium (PSB) *Bacillus megaterium* var. *phosphaticum*), singly or in combination, on the performance of blackgram cv. T-9 were studied by Tanwar *et al.* (2002) in Udaipur, Rajasthan, India during the kharif season of 1996. P as diammonium phosphate was applied during sowing. The application of 60 kg P_2O_5 ha^{-1} increased the dry matter production $plant^{-1}$ at 60 days after sowing (DAS) and at harvest, and number of primary branches $plant^{-1}$ by 52.8, 18.8 and 37.9% over the control, respectively. Seed inoculation with *Rhizobium* and PSB either singly or in combination enhanced dry matter production, number of branches $plant^{-1}$, number of nodules $plant^{-1}$, number of pods $plant^{-1}$ and number of seeds pod^{-1}. However, the inoculation of both biofertilizers was more effective than the inoculation of either of the biofertilizers. *Rhizobium* and PSB were equally effective when applied alone. The application of 40 and 60 kg P_2O_5 ha^{-1} resulted in the highest number of pods $plant^{-1}$. The highest grain, straw and biological yields were obtained with 60 kg P_2O_5 ha^{-1} (39.0, 29.7 and 31.28% increase over the control, respectively). The interaction between P rate and biofertilizers was significant with regard to the number of nodules and seed yield. The inoculation of both biofertilizers along with the application of 60 kg P_2O_5 gave the highest number of nodules $plant^{-1}$ (40.5) and seed yield.

Malik *et al.* (2002) studied the effects of seed inoculation with *Rhizobium* and P application (at 0, 30, 50, 90 and 110 kg ha^{-1}) on the growth, seed yield and quality of mungbean cv. NM-98 in a field experiment conducted at Faisalabad in Pakistan during the autumn of 2000. Plant height at harvest was highest when inoculated with *Bradyrhizobium* (68.13 cm).

Srinivas and Shaik (2002) studied the effects of N (0, 20, 40 and 60 kg ha^{-1}) and P (0, 25, 50 and 75 kg ha^{-1}) along with seed inoculation with *Rhizobium* culture on the growth and yield components of greengram were determined in a field experiment conducted during the kharif seasons. Plant height generally increased with increasing rates of P and with increasing rates of N up to 40 kg ha^{-1} followed by decrease with further increase in N. Number of seeds per pods, 1000-seed weight, seed and haulm yields generally increased. Seed inoculation with *Rhizobium* resulted in higher values for the parameters measured relative to the control. The interactions effects between N and P were not significant for the number pods plant^{-1}, pod length and seed haulm yield.

Tomar *et al.* (2003) conducted an experiment in Ujhani, Uttar Pradesh, India, during 2001-02 to study the effect of *Rhizobium*, vesicular arbuscular mycorrhiza (VAM) and phosphate solubilizing bacteria (PSB) inoculations, with different phosphorus (P) rates (0, 20 and 40 kg P ha^{-1}) on blackgram (*Vigna mungo*) and their residual effects on the growth and yield of the succeeding wheat crop. P application and inoculation treatments increased the nodulation, plant biomass, grain and straw yields, N and P content in blackgram. Residual effects of inoculation with *Rhizobium*, VAM and PSB individually or in combination and phosphorus applied to blackgram were significant on wheat. Plant dry weight, grain and straw yields and N and P uptake by wheat increased significantly. *Rhizobium* combined with VAM or PSB recorded a 10.0% increase in plant dry weight than *Rhizobium* alone. The highest residual effects on all the parameters including the wheat grain yield were recorded with combined inoculation of *Rhizobium* + VAM + PSB.

A pot study was conducted by Sheikh *et al.* (2003) to investigate the effects of seed inoculation with *Rhizobium* sp. and N:P:K (at 0:0:0, 5:15:10, 10:30:20, 15:45:30 and 20:60:40 kg ha^{-1}) growth and physiomorphological traits of blackgram cv. PU 19 irrigated with thermal power plant waste water (TPPW), sewage waste water (SW) and ground water (GW). Sampling was conducted at the vegetative (25 days after sowing, DAS), flowering (40 DAS), and fruiting (55 DAS) stages. Both SW and TPPW were efficient for improving the yield, vegetative growth, physiological characteristics and yield. Waste water application increased the leaf nitrate reductase activity, carbonic anhydrase [carbonate anhydrase] activity and total chlorophyll content of *Rhizobium*-inoculated plants. The optimum fertilizer treatment was N: P: K at 10:30:20 kg ha^{-1} and NPK-treated plants showed better performance under inoculated conditions than uninoculated.

Tanwar *et al.* (2003a) conducted a field experiment at Rajasthan College of Agriculture, Udaipur, Rajasthan, India, during the kharif season of 1996 to study the effects of P rate (0, 20, 40 or 60 kg P_2O_5 ha^{-1}) and biofertilizer (seed inoculation of *Rhizobium*, phosphate-solubilizing bacterium *Bacillus megaterium* var. *phosphaticum*, or their combination) on the performance of blackgram cv. T-9. P as diammonium phosphate was applied during sowing. The seed yield increased with the increase in P rate up to 60 kg ha^{-1}. The application of 60 kg P ha^{-1}, along with inoculation of *Rhizobium* and phosphate-solubilizing bacterium gave the highest seed yield (10.93 quintal ha^{-1}), gross return (20 244 rupees ha^{-1}), net return (15 152.5 rupees ha^{-1}), and benefit cost ratio (2.98).

Tanwar *et al.* (2003b) conducted a field study in Rajasthan, India during kharif 1996 to investigate the effect of P (0, 20, 40 and 60 kg ha^{-1}) and biofertilizers (*Rhizobium* sp. and *Bacillus megaterium* var. *phosphaticum*) on the nutrient content and uptake of blackgram. The biofertilizers were applied singly or in combination. The crop yield, N and P contents, and N and P uptake increased with increasing P rate up to 80 kg ha^{-1}. Inoculation with the combination of the biofertilizers resulted in higher yield, N and P content, N and P uptake of the grain and straw compared to no inoculation and individual inoculation.

Chapter 3
Materials & Methods

CHAPTER III

MATERIALS AND METHODS

This chapter deals with the experimental aspect of the work. Details of the experimental materials and methods followed in the study are presented in this chapter. The experiment was carried out during the period from September to December, 2007. This chapter offers a brief description of soil, treatments, design, fertilizer, biofertilizer, intercultural operations, chemical and statistical analysis.

3.1 Experimental site

The experiment was carried out at the Net House Laboratory of Soil Science Division in Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur. The experimental site is situated at 24.09^0 North Latitude and 90.50^0 'East Longitude. The elevation of the experimental site is 8.2 m above the sea level. The area belongs to the Agro-ecological Zone (AEZ 28): Madhupur Tract.

3.2 Collection and preparation of soil sample

The soil used in this experiment was collected from a selected area of Central Farm of Bangladesh Agricultural Research Institute, Joydebpur, Gazipur. The initial surface soils of 0-15 cm depth were collected. The samples were put into polyethylene bag and were taken to the laboratory. Then, it was spread on the floor and sun dried for one month. When it dried, the clods were broken with hammer to make it friable. The soil was sieved to remove weeds, stubbles and hard clods. A composite soil sample was kept in polyethylene bag for chemical analysis.

3.3 Soil

The experiment was conducted on clay loam soil of the Order Inceptisols. The soil of BARI farm is high land having irrigation facilities. The morphological, physical and chemical characteristics of the experimental soil are presented in Tables 3.1 and 3.2.

Table 3.1. Morphological characteristics of the experiment field

Characters	
General soil type	Shallow Grey Terrace Soil
Taxonomic soil classification:	
Order	Inceptisols
Sub-order	Aquept
Sub-group	Aeric Albaquept
Soil series	Chhiata

3.4 Climate

The climate of the experimental site is sub-tropical, wet and humid. Heavy rainfall occurs in the monsoon (Mid April to Mid August) and scanty during rest of the year.

Table 3.2. Physical and chemical characteristics of the soils

Characteristics	BARI farm
Mechanical fractions:	
% Sand (0.2-0.02 mm)	27.5
% Silt (0.02-0.002 mm)	33.5
%Clay (< 0.002 mm)	39.0
Textural class	Clay loam
Colour	Grey
Consistency	Sticky and mud when wet
pH (1:2.5 Soil-Water)	6.3
CEC (cmol kg^{-1})	17.5
Exchangeable K (cmol kg^{-1})	0.22
Exchangeable Ca (cmol kg^{-1})	9.41
Exchangeable Mg (cmol kg^{-1})	7.15
Exchangeable Na (cmol kg^{-1})	0.15
Organic C (%)	0.95
Total N (%)	0.072
Available P (mg kg^{-1})	13.0
Available S (mg kg^{-1})	15.0
Available Zn (mg kg^{-1})	1.59
Available Cu (mg kg^{-1})	0.59
Available Fe (mg kg^{-1})	17.9
Available Mn (mg kg^{-1})	3.5

3.5 Crop: Blackgram (*Vigna mungo* L.)

3.5.1 Blackgram variety : BARI Mash-1

BARI Mash-1

BARI Mash-1 was developed by Bangladesh Agricultural Research Institute (BARI) and it was released in 1990 by the National Seed Board. Plant height of this variety ranges from 33-35 cm; maximum field duration from 70-75 days and average yield from 1400-1500 kg ha^{-1}. In seedling stage the seedling is erect in nature; stem and leaf are dark green. Stems are comparatively bulky and leaves are broad and long. The colours of the flowers are blue and seeds are spotted light blackish brown. It is resistant to powdery mildew and downy mildew (Afzal *et al*., 1999).

3.6 Sowing period: 2007 (Kharif II)

3.7 Treatments of the experiment

The experiment consisted of two factors:

Factor A: ***Bradyrhizobium* : 2**

i) U: Without *Bradyrhizobium* inoculation

ii) I: With *Bradyrhizobium* inoculaiton

Factor B: **Five levels of phosphatic (TSP) fertilizers:**

i) P_0: 0 kg P ha^{-1}

ii) P_0: 10 kg P ha^{-1}

iii) P_{20}: 20 kg P ha^{-1}

iv) P_{30}: 30 kg P ha^{-1}

v) P_{40}: 40 kg P ha^{-1}

3.8 Treatment combinations and experimental design

Thus, there were 10 treatment combinations (5 levels of P x 2 *Bradyrhizobium*) for the experiment. The experiment was laid out in a factorial randomized complete block design (RCBD) with four replications and total treatment combinations were 40. The combinations were as follows:

Treatment combinations: *Bradyrhizobium* 2 x phosphorus levels 5 = 10

T_1: P_0 x U

T_2: P_0 x I

T_3: P_{10} x U

T_4: P_{10} x I

T_5: P_{20} x U

T_6: P_{20} x I

T_7: P_{30} x U

T_8: P_{30} x I

T_9: P_{40} x U

T_{10}: P_{40} x I

3.9 Pot preparation

To conduct the experiment earthen pots (35 x 25 cm^2) were collected and each pot was poured with 10 kg finely ground sieved soil. Triple super phosphate (TSP) was applied as recommended levels at the time of pot filling.

3.10 Fertilizer application

Muriate of potash and gypsum were applied according to the fertilizer recommendation guide (BARC, 2005) and the doses were 42 K kg ha^{-1} and 20 kg S ha^{-1}. Triple super phosphate was applied as per treatments of the experiment as recommended levels (P_0, P_{10}, P_{20}, P_{30} and P_{40} kg ha^{-1}).

3.11 Preparation and amendment of peat material

The peat soil was collected from Gopalgonj and the pH was measured by glass electrode method. The pH of the peat soil was 4.5 and it was adjusted to 6.8 by adding $CaCO_3$ solution. Fifty grams of amended peat having 8 percent moisture was taken in each polyethylene bag and the bags were sealed up. Then they were sterilized by autoclaving for three consecutive days for one hour each day. The sealed peat was ready for inoculation.

3.12 Inoculum preparation

The bradyrhizobial inoculant was prepared in the Soil Microbiology Laboratory of Bangladesh Agricultural Research Institute (BARI) using the broth culture. The *Bradyrhizobium* strain (BARI RVm-301) was collected from the stock culture of the laboratory. Yeast mannitol broth was prepared in a 500 ml Erlenmeyer flask. The liquid medium was sterilized for 30 minutes at 121^{0} C at 15 PSI. The medium was kept for cooling. After cooling, a small portion of *Bradyrhizobium* culture was aseptically transferred from agar slant to the liquid medium in the flask with the help of a sterile inoculation needle. The flask was then placed in the shaker at 28^{0}C under 120 rpm to enhance bradyrhizobial growth. After 4-5 days, the medium in the flask showed dense growth and then the broth culture was taken out from the shaker. From this ready broth, 30 ml were taken out by sterile syringe and injected into the polyethylene packet having the sterile peat. Finally, the moisture percent of the packet was adjusted to 50 percent. The inoculated packets were then incubated at 28^{0}C for two weeks to make them ready for seed inoculation.

3.13 Viability count of *Bradyrhizobium*

Viability count of bradyrhizobia in the inoculant was made one day before injecting into the peat following plate count method (Vincent, 1970). The average number of bradyrhizobia was approximately above 10^{8} cells g^{-1} in the inoculant.

3.14 Procedure for inoculation

Inoculation was done just before sowing. Healthy blackgram seeds @ 55 g for each plot were taken into polyethylene bags separately and 2 ml of the sticker solution (4% gum acacia solution) was added to each bag with sterilized pipettes. It was followed by addition of 3 g of the desired peat based *Bradyrhizobium* inoculant to each polyethylene bag and mixed thoroughly for uniform distribution and good adherence of inoculant on the surface of each seed.

3.15 Sowing

Twenty healthy blackgram seeds (BARI Mash-1), inoculated or non-inoculated, were sown on 5th September 2007 in each pot by dibbling method. After sowing the seed, the soil was saturated with water.

3.16 Intercultural operation

The seedlings of the crop emerged out within 3-4 DAS. Thinning and first weeding were done 5 days after sowing of seeds. In thinning one of the seedlings was removed from each hole in which both the planted seeds germinated. The growing points of the seedlings to be removed were pinched off. Uprooting was not done since this injures the adjacent seedlings that were left behind. Finally three plants were kept in each pot for final harvesting of the crop. Second weeding was done 20 days after the first weeding. Necessary water was added to the pots at a regular interval of 7 days until crop maturity to maintain proper moisture content. Pest did not infest the blackgram crop. No disease was observed in the pot experiment.

3.17 Collection of samples

The following observations were made regarding the growth, yield and nutrient content from the sample plants during the course of experiment.

3.17.1 Plant

Plant samples were collected at 35 and 50 DAS days intervals to record data on nodule and shoot parameters. Three plants from each pot were selected randomly and uprooted carefully by digging soil with the help of "khurpi". All possible precautions were taken to minimize the loss of nodules.

3.17.2 Study on nodulation

The plants uprooted for sampling were washed in running water cautiously to make them free from adhering soil particles and dipped in fresh water contained in a tray to avoid shrinkage of nodules. The nodules were counted, kept separately pot-wise and their dry weights were recorded. The data on nodule number and nodule mass were recorded by taking 3 randomly selected plants from each pot at different DAS. The data on nodule mass were expressed in mg $plant^{-1}$ on oven dry basis.

3.17.3 Shoot weight and root weight

After separation of the roots, dry shoot and root weights of three selected plants were recorded.

3.17.4 Shoot length and root length

Shoot and root lengths of the plant samples of three selected plants were recorded

3.18 Harvesting and data recording on yield and yield contributing characters

Yield data were collected from each pot. The seeds and stover were dried and weighed adjusting at 14% moisture content and yields were converted to g $plant^{-1}$. The following parameters were recorded:

i) Pod yield (g $plant^{-1}$)
ii) Seed yield (g $plant^{-1}$)
iii) Stover yield (g $plant^{-1}$)
iv) Plant height (cm)
v) Pot length (cm)
vi) Number of pods $plant^{-1}$
vii) Number of seeds pod^{-1}
viii) 100-seed weight (g)

3.19 Estimation of N

The N concentrations in seed were determined by micro-Kjeldahl method.

3.20 Plant analysis

3.20.1 Collection and preparation of plant samples (seeds) for chemical analysis

Plant sample (seed) was collected from bulk harvest. The seed sample was then oven dried at 65°C for 24 hours. To obtain homogenous powder, the samples were finely ground and passed through a 60-mesh sieve. The samples were stored in polyethylene bags for N and P determination.

3.20.2 Chemical analysis of plant samples

Seed of blackgram was analyzed for determination of N and P concentrations following the methods described below:

Nitrogen

The plant sample (0.1 g grain seed) was digested with conc. H_2SO_4, hydrogen peroxide and K_2SO_4-catalyst mixture (K_2SO_4: $CuSO_4$. $5H_2O$: Se = 10: 1: 0.1) at 200°C for one and a half-hour.

3.21 Nitrogen and phosphorus uptake

Nitrogen and phosphorus uptake by blackgram were computed from the respective chemical concentration and dry matter yields.

3.22 Soil analysis

Methods of soil analysis are presented in Table 3.3

3.23 Calculation of protein concentration and protein yield

Protein concentration of blackgram seed was determined by multiplying the concentration of nitrogen in blackgram seed with 6.25.

Protein yield by blackgram seed was competed from protein concentration of seed and seed yields.

Table 3.3. Methods used for soil analysis

Soil Properties	Methods
Soil texture	Hydrometer method (Black, 1965). The texture class was determined using Marshall's Triangular Coordinates of USDA system
pH	Glass-electrode pH meter with 1:2.5 soil-water ratio (Jackson, 1973).
Organic carbon	Wet digestion method (Nelson and Sommers, 1982). The organic matter was oxidized by 1N potassium dichromate and the amount of organic carbon in the aliquot was determined by titration against 0.5N ferrous sulphate heptahydrate solution in presence of 0.025 M O-phenanthroline ferrous complex.
Total N	Microkjeldahl method (Bremner and Mulvaney, 1982). Soil sample was digested with conc. H_2SO_4 in presence of K_2SO_4 catalyst mixture (K_2SO_4: $CuSO_4$: Se = 10:1:1). Nitrogen in the digest was estimated by distilling the digest with 10N NaOH followed by titration of the distillate trapped in H_3BO_3 indicator solution with 0.01N H_2SO_4.
NH_4^+-N	Extracted by 2M KCl solution (1:10 soil-extractant ratio). The aliquot was steam distilled with MgO and Devardas alloy (Keeney and Nelson, 1982).
CEC	Sodium acetate saturation method (Rhoades, 1982). The soil was leached with an excess of 1 M sodium acetate solution to remove the exchangeable cations and saturate the exchange material with sodium. The replaced sodium was determined by flame photometer.
Available P	Extracted by 0.5M $NaHCO_3$ (pH 8.5) and determined colorimetrically using molybdate blue ascorbic acid method (Olsen and Sommers, 1982).
Available K	Extracted by repeated shaking and centrifugation of the soil with neutral 1M NH_4OAc followed by decantation. The K concentration in the extract was determined by flame photometer as outlined by Knudsen *et al.* (1982).
Available S	Extracted by 500 ppm P solution from Ca (H_2PO_4).H_2O and estimated by turbidimetric method using $BaCl_2$ (Fox *et al.*, 1964).
Available Zn	Extracted by 0.05N HCl solution and determined directly by AAS (Page *et al.*, 1982).
Available Cu, Mn and Fe	Extracted by 0.005M DTPA solution and directly measured by AAS (Lindsay and Norvell, 1978).
Bulk density	Core sampling procedure (Black, 1965).
Water holding capacity	Determined gravimetrically using brass box following the method of Klute as described by Black (1965).

3.24 Statistical analysis

The collected data were analyzed statistically and Duncan's Multiple Range Test (DMRT) using a computer IRRISTAT and M-stat package programmes (Freed, 1992) adjudged the means. The correlation co-efficient and regression analysis were done for different variables wherever needed using Microsoft EXCEL programme 1997.

Chapter 4
Results and Discussion

CHAPTER IV

RESULTS AND DISCUSSION

The present experiment was carried out in order to find out the effects of different levels of phosphorus and *Bradyrhizobium* inoculant on nodulation, plant growth and yield of blackgram.

The effect of different levels of phosphorus fertilizer and *Bradyrhizobium* inoculation on nodule number, nodule weight, root and shoot weight, root and shoot length, plant height, seed, pod and stover yields, the yield contributing characters, N and P content in seed N and P uptake by seed, protein content in seed and protein yield by seed have been presented in different tables and figures and discussed in this chapter.

4.1 Nodule number on main root

4.1.1 Effect of phosphorus

Phosphorus application increased nodule number on main root appreciably at two stages of growth which became statistically significant at 5% level of probability at both 35 and 50 days after sowing (DAS) (Table 4.1). Application of P @ 20 kg ha^{-1} produced the highest number of nodules (7.12 $plant^{-1}$ at 35 and 10.12 $plant^{-1}$ and at 50 DAS) which was significantly higher over all other P levels at 35 DAS and identical to P_{10} and P_{30} at 50 DAS. Treatments P_{30}, P_{10} and P_0 were statistically identical in respect of nodule number at 35 DAS. For same parameter, treatments P_{40} and P_0 were statistically similar at 50 DAS. The lowest number of nodules (4.00 $plant^{-1}$) was recorded in P_{40} at 35 DAS and that of 6.17 nodules $plant^{-1}$ in P_0 at 50 DAS. The results indicated that number of nodules on main root increased up to P_{20} level of phosphorus and then decreased. Tomar *et al.* (2001) reported that phosphorus application influenced nodulation in blackgram

Table 4.1. Effect of phosphorus on nodulation in blackgram

P level (kg ha^{-1})	35 DAS				50 DAS			
	Main root nodule $plant^{-1}$	Branch root nodule $plant^{-1}$	Total nodule $plant^{-1}$	Nodule weight (mg $plant^{-1}$)	Main root nodule $plant^{-1}$	Branch root nodule $plant^{-1}$	Total nodule $plant^{-1}$	Nodule weight (mg $plant^{-1}$)
P_0	4.83bc	5.67b	10.50bc	13.75c	6.17b	7.02b	13.19c	38.35e
P_{10}	5.10b	6.65b	11.75b	16.56b	8.60a	10.15a	18.75b	52.50c
P_{20}	7.12a	8.88a	16.00a	19.25a	10.12a	11.88a	22.00a	72.50a
P_{30}	5.35b	6.65b	12.00b	17.60ab	9.10a	10.40a	19.50ab	57.50b
P_{40}	4.00c	5.25b	9.25c	14.58c	6.70b	7.47b	14.17c	47.10d
SE (±)	0.29	0.61	0.56	0.60	0.53	0.69	0.94	1.23

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT

4.1.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inouclant significantly increased nodule number on main root at 5% level of probability at both 35 and 50 DAS (Table 4.2). Inoculated plants produced maximum number of nodules on main root (5.90 and 9.93 $plant^{-1}$ at 35 and 50 DAS, respectively), while uninoculated plant produced minimum number of nodules on main root (4.66 and 6.35 $plant^{-1}$ at 35 and 50 DAS, respectively). Jayakumar *et al.* (1997) reported that *Rhizobium* inoculation increased number of nodules $plant^{-1}$ compared to controls. Nagarajan and Balachandar (2001) reported that seed inoculation of *Rhizobium* enhanced root nodulation.

Table 4.2. Effect of bradyrhizobial inoculant on nodulation in blackgram

Inoculant	35 DAS				50 DAS			
	Main root nodule plant^{-1}	Branch root nodule plant^{-1}	Total nodule plant^{-1}	Nodule weight (mg plant^{-1})	Main root nodule plant^{-1}	Branch root nodule plant^{-1}	Total nodule plant^{-1}	Nodule weight (mg plant^{-1})
Non-inoculated	4.66b	5.84b	10.50b	14.01b	6.35b	7.53b	13.88b	42.88b
Inoculated	5.90a	7.40a	13.30a	18.69a	9.93a	11.24a	21.17a	64.30a
SE (±)	0.18	0.39	0.35	0.38	0.33	0.44	0.60	0.78

In a column, means followed by different letter are significantly differed at 5% level by DMRT

4.1.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus and *Bradyrhizobium* inoculant application on number of nodules on main root was statistically insignificant at both 35 and 50 DAS (Table 4.3). *Bradyrhizobium* inoculant when applied with P @ 20 kg ha^{-1} produced the highest number of nodules on main root (7.72 plant^{-1} at 35 DAS and 13.10 plant^{-1} at 50 DAS) which were superior to other treatment combination. Treatment P_0 x without *Bradyrhizobium* inoculant (U) produced the lowest number of nodules (3.78 plant^{-1} at 35 and 5.10 plant^{-1} at 50 DAS). Tanwar *et al*. (2002) reported that the interaction between P rate and biofertilizers was significant in regard to the number of nodules. The inoculation of both biofertilizers along with the application of 60 kg P_2O_5 gave the highest number of nodules plant^{-1} (40.5). Tomar *et al*. (2003) observed that P application and inoculation treatments increased the nodulation in blackgram.

Table 4.3. Interaction effect of phosphorus and bradyrhizobial inoculant on nodulation in blackgram

Phosphorus x Inoculant	35 DAS				50 DAS			
	Main root nodule $plant^{-1}$	Branch root nodule $plant^{-1}$	Total nodule $plant^{-1}$	Nodule dry weight (mg $plant^{-1}$)	Main root nodule $plant^{-1}$	Branch root nodule $plant^{-1}$	Total nodule $plant^{-1}$	Nodule dry weight (mg $plant^{-1}$)
P_0 x U	3.78	4.22	8.00e	12.50	5.10	5.53	10.63	32.70f
P_0 x I	5.88	7.13	13.00bc	15.00	7.25	8.50	15.75	44.00e
P_{10} x U	4.25	5.25	9.50d	13.49	7.00	8.00	15.00	45.00e
P_{10} x I	5.95	8.05	14.00b	19.64	10.20	12.30	22.50	60.00c
P_{20} x U	6.52	8.48	15.00ab	16.00	7.15	9.10	16.25	55.00d
P_{20} x I	7.72	9.28	17.00a	22.50	13.10	14.65	27.75	90.00a
P_{30} x U	4.85	6.15	11.00cd	15.06	7.10	8.40	12.50	45.00e
P_{30} x I	5.85	7.15	13.00bc	20.14	11.10	12.40	23.50	70.00b
P_{40} x U	3.90	5.10	9.00de	13.00	5.40	6.60	12.00	36.70f
P_{40} x I	4.10	5.40	9.50d	16.15	8.00	8.33	16.33	57.50cd
SE (±)	NS	NS	0.79	NS	NS	NS	NS	1.74
CV (%)	15.5	25.7	13.3	10.5	18.2	20.9	15.5	6.5

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT
NS = Not significant
U: Non-inoculated, I: Inoculated
P_0: 0 kg P ha^{-1}, P_{10}: 10 kg P ha^{-1}, P_{20}: 20 kg P ha^{-1}, P_{30}: 30 kg P ha^{-1}, P_{40}: 40 kg P ha^{-1}

4.2 Nodule number on branch root

4.2.1 Effect of phosphorus

Phosphorus application increased nodule number on branch roots appreciably at both two stages of growth (Table 4.1). Application of P @ 20 kg ha^{-1} produced the highest number of nodules on branch root (8.88 and 11.88 $plant^{-1}$ at both 35 and 50 DAS, respectively). Treatments P_0, P_{10}, P_{30} and P_{40} were statistically identical at 35 DAS. Treatments P_0 and P_{40} were statistically similar at 50 DAS. The lowest number of branch root nodules (5.25 $plant^{-1}$) was observed in P_{40} at 35 DAS while 7.02 nodules $plant^{-1}$ was observed in P_0 at 50 DAS. Tomar *et al.* (2001) reported that phosphorus application increased nodulation in blackgram.

4.2.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inouclant significantly increased nodule number on branch root at 5% level of probability at both 35 and 50 DAS (Table 4.2). Inoculated plants produced maximum number of nodules (7.40 and 11.24 $plant^{-1}$ at 35 and 50 DAS, respectively). Uninoculated plant produced minimum number of branch root nodules at both stages. The above result was similar with the result at Bhuiyan *et al.* (2006). Jayakumar *et al.* (1997) reported that *Rhizobium* inoculation increased number of nodules $plant^{-1}$ compared to control. Nagarajan and Balachandar (2001) reported that seed inoculation of *Rhizobium* enhanced root nodulation.

4.2.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus and *Bradyrhizobium* inoculant application on number of nodules on branch root was statistically insignificant at both 35 and 50 DAS (Table 4.3). Both at 35 and 50 DAS, *Bradyrhizobium* inoculant when applied with P @ 20 kg ha^{-1} produced the highest number of nodules on branch root (9.28 and 14.65 $plant^{-1}$) which were superior to other treatments. P_0 x U treatment combination produced the lowest number of nodules (4.22 and 5.53 $plant^{-1}$ at 35 and 50 DAS, respectively). Tanwar *et al.* (2002) reported that the interaction between P rate and biofertilizers was significant with regard to the number of nodules. The inoculation of both biofertilizers along with the application of 60 kg P_2O_5 gave the highest number of nodules $plant^{-1}$ (40.5). Tomar *et al.* (2003) observed that P application and inoculation treatments increased the nodulation in blackgram.

4.3 Total number of nodule

4.3.1 Effect of phosphorus

Phosphorus application increased total nodule number appreciably at two stages of growth which became statistically significant at 5% level of probability (Table 4.1 and Figure 4.1). Application of P @ 20 kg ha^{-1} produced the highest number of nodules (16 $plant^{-1}$) at 35 DAS and that of 22 nodules $plant^{-1}$ at 50 DAS. In case of 35 DAS, treatments P_{30}, P_{10} and P_0 were statistically similar. At 50 DAS, treatment P_{40} and P_0 were statistically identical. Tomar *et al.* (2001) found that phosphorus application increased nodulation in blackgram.

4.3.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculant significantly increased total nodule number at 5% level of probability at 35 and 50 DAS (Table 4.2 & Figure 4.2). Inoculated plants produced maximum number of total nodules (13.30 and 21.17 $plant^{-1}$ at 35 and 50 DAS, respectively) but uninoculated plants produced minimum number of nodules (10.50 and 13.88 $plant^{-1}$ at 35 and 50 DAS, respectively). The above result was similar with the results obtained by Bhuiyan *et al.* (2005) and Bhuiyan *et al.* (2006, 2007a). Jayakumar *et al.* (1997) reported that *Rhizobium* inoculation increased number of nodules $plant^{-1}$ compared to controls. Sharma *et al.* (1999) found that application of inoculation increased nodulation in blackgram. Nagarajan and Balachandar (2001) reported that seed inoculation of *Rhizobium* enhanced root nodulation. They reported that *Rhizobium* gave the highest nodule number (23.3 and 24.0) in blackgram and greengram. Bhattacharyya and Pal (2001a) reported that *Rhizobium* inoculation significantly influenced the number of nodules $plant^{-1}$. Tomar *et al.* (2001) found that *Rhizobium* gave the highest and 21.0% more nodules compared to uninoculated control. Singha and Sarma (2001) reported that *Rhizobium* inoculation significantly increased the nodule number compared to the control indicating increased efficiency of the crop to fix the atmospheric N. Reddy and Mallaiah (2001) found that a maximum of 84 nodules $plant^{-1}$ were found due to *Rhizobium* inoculation during the reproductive stage of the crop. Chatterjee and

Bhattacharjee (2002) noted that plants inoculated with *Bradyrhizobium* strains showed an increased rate of nodulation. Kumari and Nair (2003) observed that the extent of root nodulation were more in blackgram and greengram where *Bradyrhizobium* inoculation was done.

4.3.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* inoculant application on total number of nodules was statistically significant at 35 DAS but insignificant at 50 DAS (Table 4.3). *Bradyrhizobium* inoculant when applied with P @ 20 kg ha^{-1} produced the highest number of nodules (17.00 $plant^{-1}$ at 35 DAS and 27.75 $plant^{-1}$ at 50 DAS). The lowest number of nodules both at 35 and 50 DAS were observed in P_0 x U treatment combination. Tanwar *et al.* (2002) reported that the interaction between P rate and biofertilizers was significant with regard to the number of nodules. The inoculation of biofertilizers along with the application of 60 kg P_2O_5 ha^{-1} gave the highest number of nodules $plant^{-1}$ (40.5). Tomar *et al.* (2003) observed that P application and inoculation treatments increased the nodulation in blackgram.

4.4 Nodule dry weight

4.4.1 Effect of phosphorus

Nodule dry weights were increased significantly at both 35 and 50 DAS by phosphorus application (Table 4.1 and Figure 4.3). Application of phosphorus @ 20 kg ha^{-1} gave the highest nodule weights (19.25 and 72.50 mg $plant^{-1}$ at 35 and 50 DAS, respectively). The lowest nodule weights (13.75 and 38.35 mg plant-1 at 35 and 50 DAS, respectively) were observed in P application @ 0 kg ha^{-1}. The results are in line with those of some authors. Tomar *et al.* (2001) who reported that application of phosphorus increased nodule weight in blackgram.

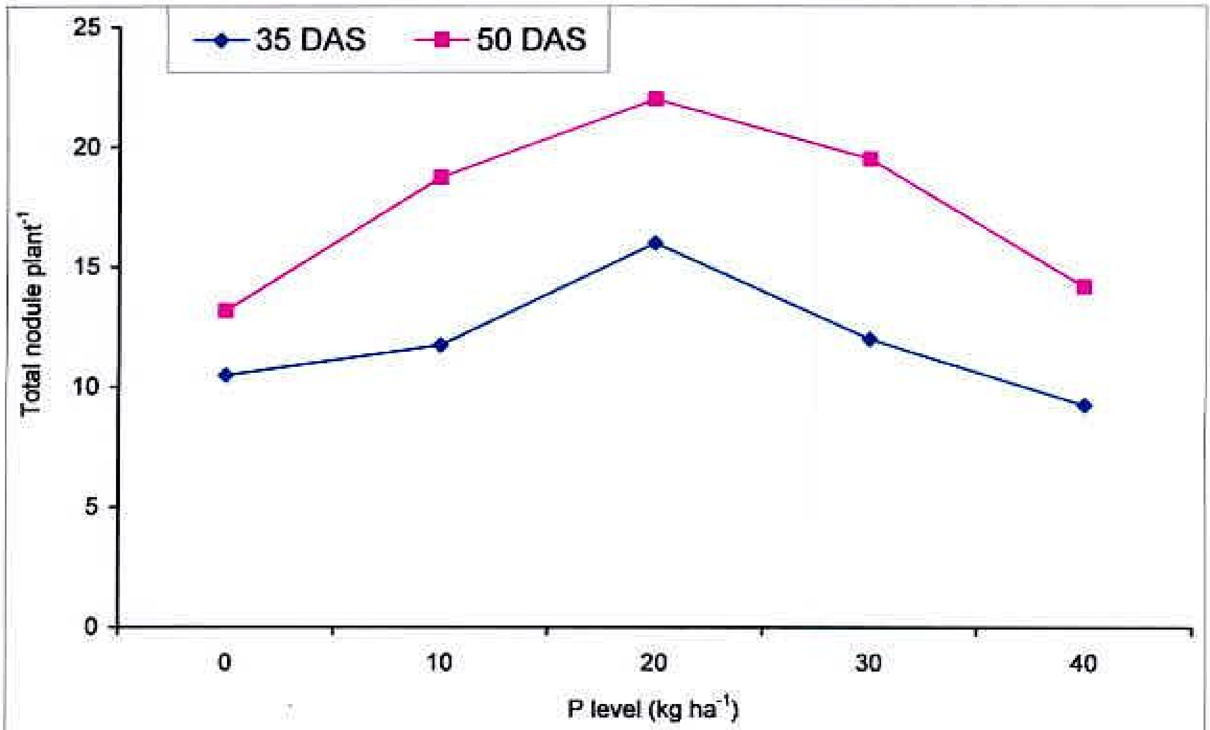

Fig. 4.1. Effect of different levels of phosphorus on total nodule number in blackgram

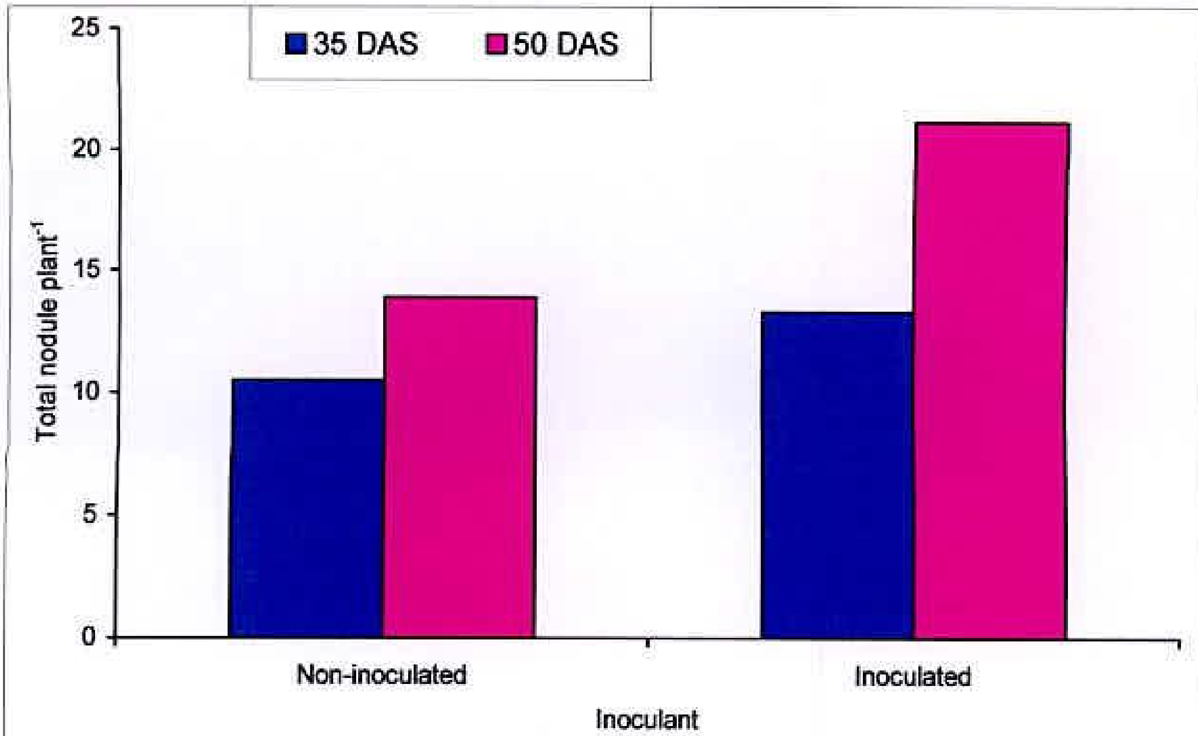

Fig. 4.2. Effect of bradyrhizobial inoculant on total nodule number in blackgram

4.4.2 Effect of *Bradyrhizobium* inoculant

The effect of *Bradyrhizobium* inoculation on nodule weight was significant at both 35 and 50 DAS (Table 4.2 and Figure 4.4). Inoculated plants produced higher nodule weights (18.69 and 64.30 mg $plant^{-1}$) at 35 and 50 DAS, respectively whereas uninoculated plants produced comparatively lower nodule weights (14.01 and 42.88 mg $plant^{-1}$ at 35 and 50 DAS, respectively). Singha and Sarma (2003) reported that *Rhizobium* inoculant had 9.5% higher nodule dry weight than non-inoculated control. Sharma *et al.* (1999) found that application of inoculants increased nodule dry weight $plant^{-1}$. Nagarajan and Balachandar (2001) reported that *Rhizobium* gave higher nodule weight (45.3 and 42.3 mg) in blackgram and greengram. Tomar *et al.* (2001) found that *Rhizobium* gave the highest and 34.7% more nodule dry mass. Kumari and Nair (2003) observed significant increases in nodule dry weight due to *Rhizobium* inoculation.

4.4.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

Phosphorus x *Bradyrhizobium* interaction effect on nodule weight was insignificant at 35 DAS but highly significant at 50 DAS (Table 4.3). At 50 DAS, *Bradyrhizobium* inoculant when applied with P @ 20 kg ha^{-1} gave the highest nodule weight (90 g $plant^{-1}$) which was superior to all other treatments. P_{20} x I treatment combination produced the highest nodule weight (22.50 and 90.00 mg $plant^{-1}$ at both 35 and 50 DAS respectively). Treatment combination P_0 x U gave the lowest nodule weight (12.50 and 32.70 at 35 and 50 DAS, respectively).

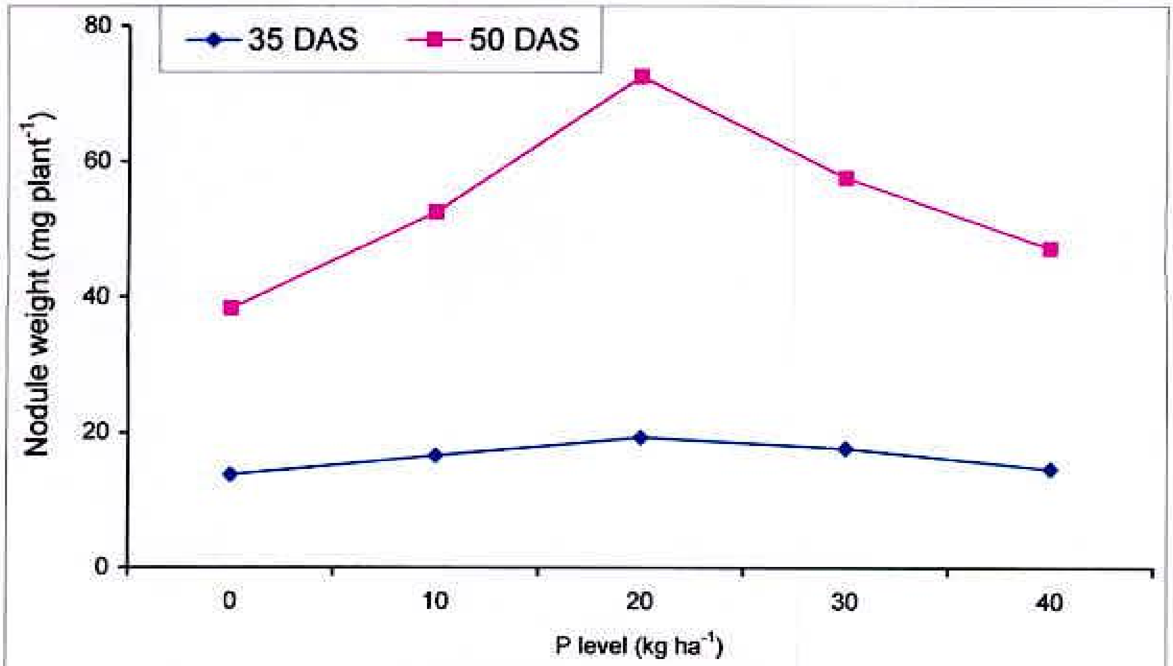

Fig. 4.3. Effect of different levels of phosphorus on nodule weight in blackgram

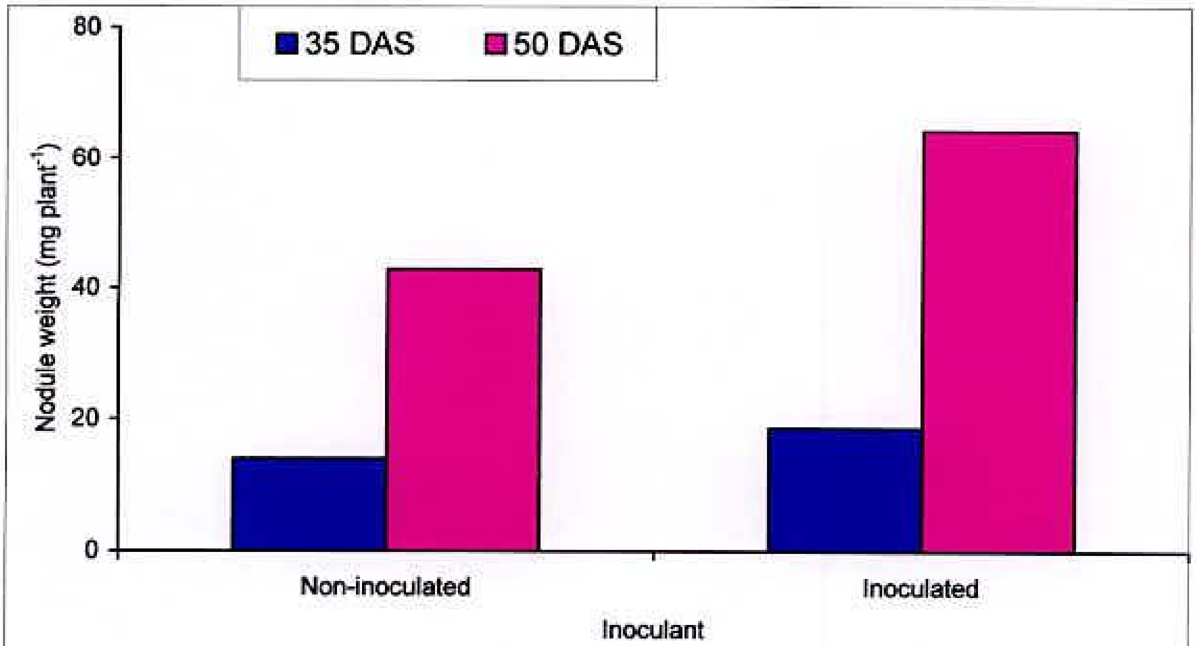

Fig. 4.4. Effect of bradyrhizobial inoculant on nodule weight in blackgram

4.5 Root weight

4.5.1 Effect of phosphorus

Root weights were statistically significant at both 35 and 50 DAS due to application of different levels of phosphorus (Table 4.4). At 35 DAS, application of phosphorus @ 20 kg ha^{-1} gave the highest root weight (0.10 g $plant^{-1}$). This was identical to P_{10} and P_{30}. Treatment P_{40} and P_0 were statistically similar. In case of 50 DAS, the highest root weight (0.39 g $plant^{-1}$) was recorded with P_{30} level and the lowest (0.21 g $plant^{-1}$) with P_0 level. Treatment P_{40} and P_{10} were statistically identical at 50 DAS. The lowest root weight was recorded in P_0 level at both 35 and 50 DAS. The results are in agreement with the findings of Singh and Singh (2004).

Table 4.4. Effect of phosphorus on weight and length of root and shoot in blackgram

P level (kg ha^{-1})	35 DAS				50 DAS			
	Root weight (g $plant^{-1}$)	Shoot weight (g $plant^{-1}$)	Root length (cm)	Shoot length (cm)	Root weight (g $plant^{-1}$)	Shoot weight (g $plant^{-1}$)	Root length (cm)	Shoot length (cm)
P_0	0.07b	1.06d	6.30	34.87d	0.21d	3.89bc	7.13	39.75b
P_{10}	0.09a	1.24c	6.94	38.80c	0.25c	4.24ab	7.38	44.45b
P_{20}	0.10a	1.55a	7.63	42.40a	0.34b	4.58a	8.50	52.50a
P_{30}	0.09a	1.36b	7.32	40.88b	0.39a	3.55cd	8.09	43.38b
P_{40}	0.07b	1.03d	7.00	35.81d	0.27c	3.02d	7.57	40.67b
SE (±)	0.004	0.03	NS	0.41	0.009	0.20	NS	1.99

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT
NS = Not significant

4.5.2 Effect of *Bradyrhizobium* inoculant

It was found from Table 4.5 that *Bradyrhizobium* had significant effect on root weight. Inoculated plants produced higher root weights (0.09 and 0.34 g $plant^{-1}$ at 35 and 50 DAS, respectively) than non-inoculated plants. Srivastav and Poi (2000) found that inoculation with M-10 strain in greengram resulted in the highest dry matter production. Sharma *et al.* (2000) reported that seed inoculated with 1 of 9 *Rhizobium* strains increased dry matter accumulation. Parveen *et al.* (2002) reported that the maximum root dry weight (0.37 g plant^{-1}) was observed with single *Bradyrhizobium* sp.

Table 4.5. Effect of bradyrhizobial inoculant on weight and length of root and shoot in blackgram

Inoculant	35 DAS				50 DAS			
	Root weight (g plant^{-1})	Shoot weight (g plant^{-1})	Root length (cm)	Shoot length (cm)	Root weight (g plant^{-1})	Shoot weight (g plant^{-1})	Root length (cm)	Shoot length (cm)
Non-inoculated	0.07b	1.09b	6.67b	35.62b	0.24b	3.31b	7.05b	39.75b
Inoculated	0.09a	1.40a	7.48a	41.48a	0.34a	4.40a	8.41a	48.55a
SE (±)	0.002	0.015	0.18	0.26	0.006	0.13	0.25	1.26

In a column, means followed by different letter are significantly differed at 5% level by DMRT

4.5.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

Interaction effect of phosphorus x *Bradyrhizobium* was insignificant on root weight at 35 DAS but significant at 50 DAS (Table 4.6). Application of phosphorus along with *Bradyrhizobium* produced the highest root weight at both 35 and 50 DAS. At 35 DAS, the treatment combination P_{20} x I and P_{10} x I produced the highest root weight (0.11 g plant^{-1}). In case of 50 DAS, the highest root weight (0.45 g plant^{-1}) was recorded with P_{30} x I treatment combination and the lowest (0.15 g plant^{-1}) was recorded with P_0 x U treatment combination. Treatment combinations P_{20} x I, P_{40} x I and P_{30} x U were statistically identical at 50 DAS. Tomar *et al.* (2003) observed that P application and inoculation treatments increased the plant biomass in blackgram.

Table 4.6. Interaction effect of phosphorus and bradyrhizobial inoculant on weight and length of root and shoot in blackgram

Phosphorus x Inoculant	35 DAS				50 DAS			
	Root weight (g plant^{-1})	Shoot weight (g plant^{-1})	Root length (cm)	Shoot length (cm)	Root weight (g plant^{-1})	Shoot weight (g plant^{-1})	Root length (cm)	Shoot length (cm)
P_0 x U	0.06	0.83f	6.00	32.25	0.15h	3.01	6.50	38.00
P_0 x I	0.07	1.29cd	7.00	37.50	0.26ef	4.76	7.75	41.50
P_{10} x U	0.07	1.17e	6.38	36.10	0.22fg	3.75	6.75	40.00
P_{10} x I	0.11	1.30cd	7.50	41.50	0.28de	4.73	8.00	48.90
P_{20} x U	0.09	1.31c	7.25	39.50	0.31cd	4.25	7.50	42.50
P_{20} x I	0.11	1.79a	8.00	45.30	0.36b	4.91	9.50	62.50
P_{30} x U	0.08	1.20de	7.13	37.50	0.32bc	3.18	7.50	40.75
P_{30} x I	0.10	1.51b	7.50	44.25	0.45a	3.91	8.67	46.00
P_{40} x U	0.06	0.93f	6.60	32.75	0.21g	2.35	7.00	37.50
P_{40} x I	0.08	1.13e	7.40	38.87	0.33bc	3.68	8.13	43.83
SE (±)	NS	0.04	NS	NS	0.013	NS	NS	NS
CV (%)	15.0	5.7	11.4	3.00	9.4	14.8	14.7	12.8

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT
NS = Not significant
U: Non-inoculated, I: Inoculated

4.6 Shoot weight

4.6.1. Effect of phosphorus

Phosphorus application increased shoot weight significantly at both 35 and 50 DAS (Table 4.4). The highest shoot weight (1.55 and 4.58 g plant^{-1} at both 35 and 50 DAS, respectively) were recorded with P_{20} level treated plot. At 35 DAS, treatments P_{20} and P_{30} were statistically alike. At 50 DAS, treatments P_{10} and P_{20} were statistically identical. The lowest shoot weights (1.03 and 3.02 g plant^{-1}) were recorded with P_{40} level at both 35 and 50

DAS, respectively. Singh and Singh (2004) found that dry matter yield increased with the application of phosphorus.

4.6.2 Effect of *Bradyrhizobium* inoculant

Effect of *Bradyrhizobium* inoculum on shoot weight was highly significant at both 35 and 50 DAS (Table 4.5). Inoculated plants produced higher shoot weight (1.40 and 4.40 g $plant^{-1}$ at 35 and 50 DAS, respectively) while uninoculated plants produced lower shoot weights (1.09 and 3.31 g $plant^{-1}$ at 35 and 50 DAS, respectively). Jayakumar *et al.* (1997) reported that *Rhizobium* inoculation increased the dry weight of plants compared to controls. Srivastav and Poi (2000) found that inoculation with M-10 strain in greengram resulted in the highest dry matter production. Sharma *et al.* (2000) reported that seed inoculated with 1 of 9 *Rhizobium* strains increased dry matter accumulation. Manivannan *et al.* (2003) reported that *Rhizobium* seed treatment produced markedly higher dry matter.

4.6.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* on shoot weight was significant at 5% level at 35 DAS but insignificant at 50 DAS (Table 4.6). At 35 DAS, the highest shoot weight (1.79 g $plant^{-1}$) was recorded when *Bradyrhizobium* inoculant was applied along with P_{20} level, which was statistically the best of all. Treatment P_0 x U produced the lowest shoot weight (0.83 g $plant^{-1}$). In case of 50 DAS, the highest shoot weight (4.91 g $plant^{-1}$) was recorded with P_{20} x I treatment combination and the lowest (2.35 g $plant^{-1}$) was recorded with P_{40} x U treatment combination. Sharma *et al.* (1993) observed that in pot experiments that stover yields of *Vigna radiata* cv. Pusa Baishakhi increased with P up to or equivalent of 60 kg P ha^{-1} and with *Bradyrhizobium* inoculation. Singha and Sarma (2001) reported that P at 45 kg ha^{-1} with *Rhizobium* inoculation produced the highest straw yield and was at par with the application of 25 and 35 kg P ha^{-1}. Tomar *et al.* (2003) observed that P application and inoculation increased the plant biomass in blackgram.

4.7 Root length

4.7.1 Effect of phosphorus

Phosphorus application had no significant effect on root length at both 35 and 50 DAS (Table 4.4). The highest root lengths (7.63 and 8.50 cm at 35 and 50 DAS, respectively) were recorded with P_{20} level in both growth stages. The lowest root lengths (6.30 and 7.13 cm at 35 and 50 DAS, respectively) were recorded with P_0 level in both the growth stages.

4.7.2 Effect of *Bradyrhizobium* inoculant

The effect of *Bradyrhizobium* inoculum on root length was significant at both 35 and 50 DAS at 5% level (Table 4.5). The inoculated plants produced the highest root lengths (7.48 and 8.41 cm at 35 and 50 DAS, respectively), while non-inoculated plants produced the lowest root lengths (6.67 and 7.05 cm at 35 and 50 DAS, respectively). The above results confirmed the results of Sudhakar *et al.* (1989) who conducted an experiment on blackgram and found that *Rhizobium* inoculation increased crop growth rate compared to non-inoculated control. Sharma *et al.* (2000) reported that plant growth was increased with *Rhizobium* inoculation, with the local strain giving the best results. Sarker *et al.* (2002) reported that bradyrhizobial strain 480-M resulted the longest roots (14.72 cm).

4.7.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect was found in-significant both at 35 and 50 DAS (Table 4.6). The treatment combination P_{20} x I gave the highest root lengths (8.00 and 9.50 cm at both 35 and 50 DAS, respectively) and the treatment combination P_0 x U gave the lowest root lengths (6.00 and 6.50 cm at both 35 and 50 DAS, respectively).

4.8 Shoot length

4.8.1 Effect of phosphorus

Effect of phosphorus on shoot length was significant at 5% level at both 35 and 50 DAS (Table 4.4). The highest shoot lengths (42.40 and 62.50 cm at 35 and 50 DAS, respectively) were recorded with P_{20} level. At 35 DAS, treatments P_{40} and P_0 were statistically identical and at 50 DAS all treatments were statistically similar except P_{20}. The lowest shoot lengths (34.87 and 39.75 cm at 35 and 50 DAS, respectively) were recorded with P_0 level. Mahmud *et al.* (1997) reported that plant height was significantly increased with increasing phosphorus application. Similar results were observed by Maqsood *et al.* (2001) and Kumar *et al.* (2000)

4.8.2 Effect of *Bradyrhizobium* inoculant

The effect of *Bradyrhizobium* inoculant on shoot length was significant at 5% level at both 35 and 50 DAS (Table 4.5). The highest shoot lengths (41.48 and 48.55 cm at 35 and 50 DAS, respectively) were observed in the inoculated plants and the lowest (35.62 and 39.75 cm at 35 and 50 DAS, respectively) in the non-inoculated control. Sharma *et al.* (2000) reported that growth increased with *Rhizobium* inoculation, with the local strain giving the best results. Nagarajan and Balachandar (2001) reported that *Rhizobium* gave the maximum plant height (42.7 and 53.7 cm for blackgram and greengram, respectively). Bhattacharya and Pal (2001) reported that *Rhizobium* inoculation significantly increased plant height. Malik *et al.* (2002) studied that plant height at harvest was the highest when inoculated with *Bradyrhizobium* (68.13 cm). Ashraf *et al.* (2003) found that the tallest plants (69.93 cm) were obtained with seed inoculation. Kumari and Nair (2003) observed that the extent of plant growth were more in blackgram and greengram where *Bradyrhizobium* inoculation was done. Sriramachandrasekharan and Vaiyapuri (2003) reported that *Rhizobium*-inoculated blackgram showed better growth than the non-inoculated crop.

4.8.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect was found insignificant at both 35 and 50 DAS (Table 4.6). The treatment combination P_{20} x I gave the highest shoot length (45.30 and 62.50 cm at both 35 and 50 DAS, respectively). At 35 DAS, the P_0 x U treatment combination gave the lowest shoot length (32.25 cm) and in case of 50 DAS, the treatment combination P_{40} x U gave the lowest shoot length (37.50 cm).

4.9 Pod yield

4.9.1 Effect of phosphorus

Effect of phosphorus on pod yield was highly significant (Table 4.7). The highest pod yield (3.31 g palnt^{-1}) was recorded with P_{20} level which was superior to all other treatments but statistically similar with P_{30} treatment. The treatments P_{40} and P_{10} were statistically identical. The lowest pod yield (2.23 g plant^{-1}) was recorded with P_0 level. Mahmud *et al.* (1997) reported that weights of pod plant^{-1} significantly increased with increasing phosphorus application. The results are in agreement with Kumar *et al.* (2000), Maqsood *et al.* (2001), Tomar *et al.* (2001), Singh *et al.* (2002), Patel and Thakur (2003), Singh and Singh (2004) and Singh (2004).

Table 4.7. Effect of phosphorus on yield of blackgram

P level (kg ha^{-1})	Pod yield (g plant^{-1})	Seed yield (g plant^{-1})	Stover yield (g plant^{-1})
P_0	2.23c	1.71d	2.43
P_{10}	2.39bc	1.92c	2.90
P_{20}	3.31a	2.64a	3.33
P_{30}	3.10a	2.20b	2.85
P_{40}	2.68b	1.84cd	2.84
SE (±)	0.13	0.07	NS

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT
NS = Not significant

4.9.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculant significantly increased pod yield at 5% level of probability (Table 4.8). Inoculated plants produced higher pod yield (3.09 g $plant^{-1}$) whereas non-inoculated plants produced lower pod yield (2.39 g $palnt^{-1}$). Srivastav and Poi (2000) found that NK-4 inoculation into blackgram resulted in the highest grain yield. Sharma *et al.* (2000) reported that yield increased with *Rhizobium* inoculation. Nagarajan and Balachandar (2001) reported that *Rhizobium* gave higher grain yield (758.3 and 732.0 kg ha^{-1}) in blackgram and greengram respectively. Malik *et al.* (2002) studied that seed inoculation with *Rhizobium* resulted in the highest seed yield (1158 kg ha^{-1}).

Table 4.8. Effect of bradyrhizobial inoculant on yield of blackgram

Inoculant	Pod yield (g $plant^{-1}$)	Seed yield (g $plant^{-1}$)	Stover yield (g $plant^{-1}$)
Non-inoculated	2.39b	1.84 b	2.65b
Inoculated	3.09a	2.28 a	3.09a
SE (±)	0.08	0.04	0.14

In a column, means followed by different letter are significantly differed at 5% level by DMRT

4.9.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* on pod yield was highly significant (Table 4.9). Inoculation along with phosphorus @ 20 kg ha^{-1} gave the highest pod yield (4.04 g $plant^{-1}$) which was superior to all other treatments and similar with P_{30} x I treatment combination. The lowest pod yield (2.10 g $plant^{-1}$) was recorded in P_0 x U treatment combination. Sharma *et al.* (1993) observed in pot experiments that seed yields of *Vigna radiata* cv. Pusa Baishakhi increased with P upto or equivalent of 60 kg P ha^{-1} and with *Bradyrhizobium* inoculation. Singha and Sarma (2001) reported that P at 45 kg ha^{-1} with *Rhizobium* inoculation produced the highest grain yield and was at par with the application of

25 and 35 kg P ha^{-1}. Tanwar *et al.* (2002) reported that the interaction between P rate and biofertilizers was significant with regard to the seed yield. The inoculation of both biofertilizers along with the application of 60 kg P_2O_5 gave the highest seed yield. Srinivas and Shaik (2002) studied that the interactions effects between N and P were not significant for seed yield. Tomar *et al.* (2003) observed that P application and inoculation treatments increased grain yields in blackgram.

Table 4.9. Interaction effect of phosphorus and bradyrhizobial inoculant on yield of blackgram

Phosphorus x Inoculant	Pod yield (g $plant^{-1}$)	Seed yield (g $plant^{-1}$)	Stover yield (g $plant^{-1}$)
P_0 x U	2.01c	1.57f	2.04
P_0 x I	2.45bc	1.85de	2.83
P_{10} x U	2.17c	1.76ef	2.48
P_{10} x I	2.61bc	2.08cd	3.32
P_{20} x U	2.58bc	2.23c	3.28
P_{20} x I	4.04a	3.04a	3.37
P_{30} x U	2.63bc	1.86de	2.61
P_{30} x I	3.57a	2.53b	3.09
P_{40} x U	2.58bc	1.79ef	2.82
P_{40} x I	2.79b	1.88de	2.86
SE (±)	0.18	0.09	NS
CV (%)	13.0	9.1	21.1

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT
NS = Not significant
U: Non-inoculated, I: Inoculated

4.10 Seed yield

4.10.1 Effect of phosphorus

The effect of phosphorus on seed yield was highly significant (Table 4.7 and Figure 4.5). The highest seed yield (2.64 g $plant^{-1}$) was recorded with P_{20} level which was significantly different from and superior to all other treatments. The treatment P_{30} gave the second higher seed yield (2.20 g $plant^{-1}$). The treatment P_0 gave the lowest seed yield (1.71 g $plant^{-1}$) which was statistically similar with P_{40} levels. Mahmud *et al.* (1997) reported that weights of pod $plant^{-1}$ were significantly increased with increasing phosphorus level. The results corroborated with the findings of Kumar *et al.* (2000), Tomar *et al.* (2001), Singh *et al.* (2002), Patel and Thakur (2003), Singh and Singh (2004) and Singh (2004).

4.10.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculant significantly increased seed yield at 5% of probability (Table 4.8 and Figure 4.6). Inoculated plants produced higher seed yield (2.28 g $plant^{-1}$) whereas non-inoculated plants produced lower seed yield (1.84 g $plant^{-1}$). The result was similar with the report of Bhuiyan *et al.* (2006). Mal ik *et al.* (2002) studied that seed inoculation with *Rhizobium* application resulted in the highest seed yield (1158 kg ha^{-1}). Chatterjee and Bhattacharjee (2002) noted that plants inoculated with *Bradyrhizobium* strains showed increased seed yield over control. Parveen *et al.* (2002) reported that the maximum seed yield (6.6 g $plant^{-1}$) were observed with single *Bradyrhizobium* sp. Osunde *et al.* (2003) reported that *Bradyrhizobium* inoculation increased 40% seed yield. Asraf *et al.* (2003) found that seed inoculation was optimum for the production of high seed yield by mungbean cv. NM-98. Manivannan *et al.* (2003) reported that *Rhizobium* seed treatment had markedly higher crop yield. Kumari and Nair (2003) observed significant increase in yield due to *Rhizobium* inoculation. Sriramachandrasekharan and Vaiyapuri (2003) reported that *Rhizobium*-inoculated blackgram showed higher pod yield (50.3 g) than the non-inoculated crop.

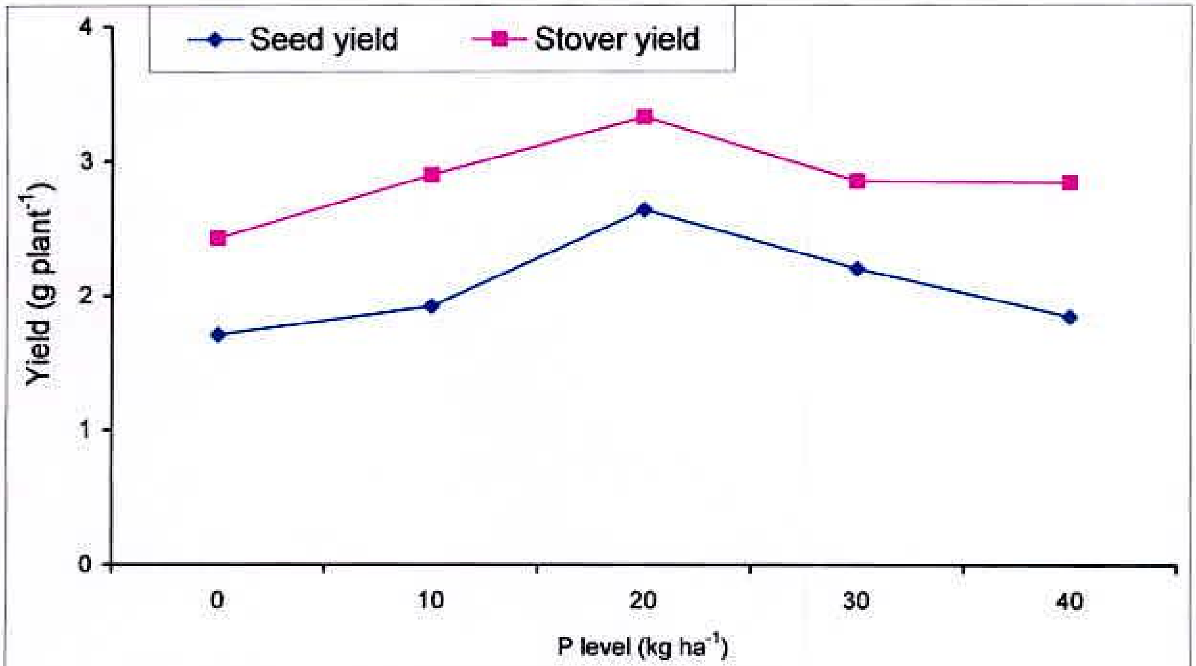

Fig. 4.5. Effect of different levels of phosphorus on seed and stover yield in blackgram

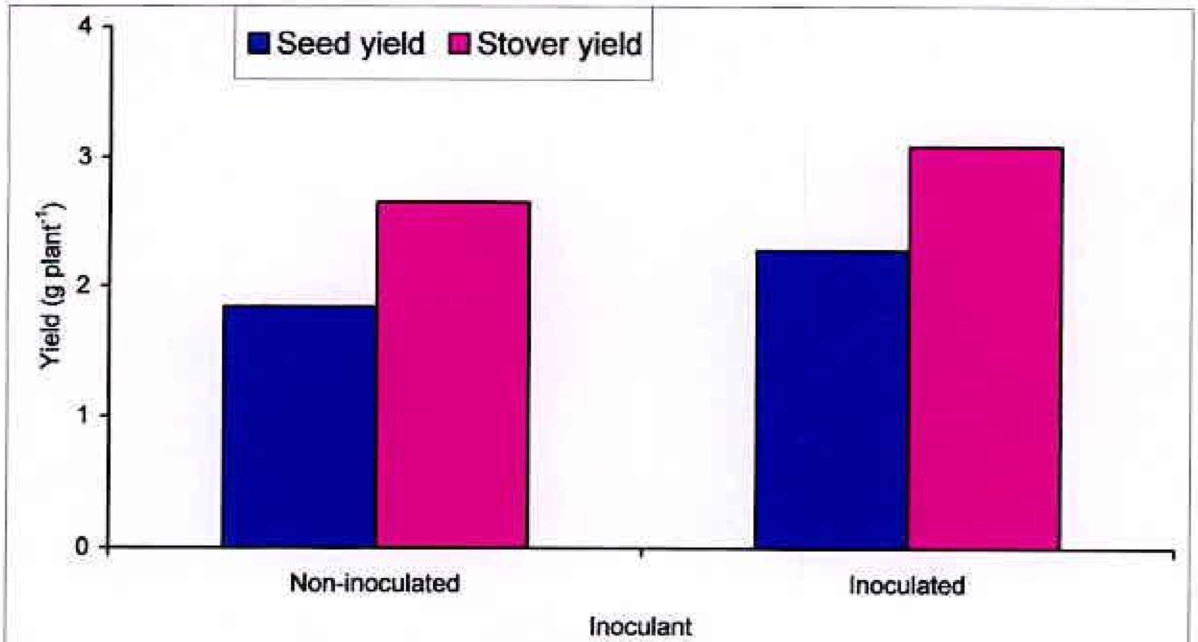

Fig. 4.6. Effect of bradyrhizobial inoculant on seed and stover yield weight in blackgram

4.10.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* on seed yield was highly significant (Table 4.9). *Bradyrhizobium* inoculation when applied with phosphorus @ 20 kg ha^{-1} gave the highest seed yield (3.04 g $plant^{-1}$) which was significantly different from and superior to all other treatments. The second highest seed yield (2.53 g $plant^{-1}$) was recorded with treatment combination of P_{30} x I level and P_0 x U gave the lowest seed yield (1.57 g $plant^{-1}$). Singha and Sarma (2001) reported that P at 45 kg ha^{-1} with *Rhizobium* inoculation produced the highest grain yield and was at par with the application of 25 and 35 kg P ha^{-1}. Tanwar *et al.* (2002) reported that the interaction between P rate and biofertilizers was significant with regard to the seed yield. The inoculation of both biofertilizers along with the application of 60 kg P_2O_5 gave the highest seed yield. Srinivas and Shaik (2002) studied that the interaction effects between N and P were not significant for seed yield. Tomar *et al.* (2003) observed that P application and inoculation treatments increased grain yields in blackgram. Tanwar *et al.* (2003a) observed that the application of 60 kg P ha^{-1} along with inoculation of *Rhizobium* gave the highest seed yield (10.93 quintal ha^{-1}).

4.11 Stover yield

4.11.1 Effect of phosphorus

Effect of phosphorus on stover yield was insignificant (Table 4.7 and Figure 4.5). The highest stover yield (3.33 g $plant^{-1}$) was recorded with P_{20} level which was superior to all other treatments. The treatment P_0 gave the lowest stover yield (2.43 g $plant^{-1}$). Similar results due to phosphorus application were observed by Ali *et al.* (1995), Trividi *et al.* (1997b), Tomar (1998), Singh and Singh (2004) and Singh (2004).

4.11.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculant significantly increased stover yield at 5% level of probability (Table 4.8 and Figure 4.6). Inoculated plants produced significantly higher stover yield (3.09 g $plant^{-1}$) than non-inoculated plants (2.65 g $plant^{-1}$). Nagarajan and Balachandar (2001) reported that seed inoculation of *Rhizobium* enhanced biomass. Srinivas and Shaik (2002) studied that seed inoculation with *Rhizobium* culture increased haulm yields relative to the control. Sriramachandrasekharan and Vaiyapuri (2003) reported that *Rhizobium*-inoculated blackgram showed higher stover yield pot^{-1} (81.1 g) than the non-inoculated crop.

4.11.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus X *Bradyrhizobium* on stover yield was insignificant (Table 4.9). *Bradyrhizobium* inoculant when applied with phosphorus @ 20 kg ha^{-1} gave the highest stover yield (3.37 g $plant^{-1}$) which was superior to all other treatments. The lowest stover yield (2.04 g $plant^{-1}$) was recorded with the treatment combination of P_0 x U level. Singha and Sarma (2001) reported that P at 45 kg ha^{-1} with *Rhizobium* inoculation produced the highest straw yield and was at par with the application of 25 and 35 kg P ha^{-1}. Srinivas and Shaik (2002) studied that the interactions effects between N and P were not significant for haulm yield. Tomar *et al.* (2003) observed that P application and inoculation treatments increased the plant biomass and yield in blackgram.

4.12 Plant height

4.12.1 Effect of phosphorus

Effect of phosphorus on plant height was insignificant (Table 4.10). The highest plant height (41.3 cm) was recorded with P_{30} level. The treatment P_0 produced the lowest plant height (37.7 cm). This indicated that phosphorus had no effect on plant height of blackgram. Mahmud *et al.* (1997) reported that phosphorus application increased plant height in blackgram. The result corroborated with the findings of Maqsood *et al.* (2001) and Kumar *et al.* (2000).

Table 4.10. Effect of phosphorus on yield contributing characters of blackgram

P level (kg ha^{-1})	Plant height (cm)	Pod length (cm)	Pods $plant^{-1}$	Seeds pod^{-1}	100-seed weight (g)
P_0	37.7	3.56	6.59b	4.71	5.16b
P_{10}	38.3	3.64	6.82b	4.83	5.30b
P_{20}	39.5	4.10	8.44a	5.03	6.02a
P_{30}	41.3	3.78	7.36ab	5.11	5.78a
P_{40}	38.6	3.51	7.42ab	4.73	5.17b
SE (±)	NS	NS	0.42	NS	0.16

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT
NS = Not significant

4.12.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculant significantly increased plant height (Table 4.11). Inoculated plants produced higher plant height (40.5 cm) than non-inoculated plants (37.6 cm). The above results revealed that *Bradyrhizobium* inoculant had significant effect on plant height of blackgram. Sharma *et al*. (2000) reported that the growth was increased with *Rhizobium* inoculation and the local strain gave the best results. Nagarajan and Balachandar (2001) reported that *Rhizobium* gave the greatest plant height (42.7 and 53.7 cm for blackgram and greengram, respectively). Bhattacharyya and Pal (2001) reported that *Rhizobium* inoculation significantly influenced plant height. Malik *et al*. (2002) studied that plant height at harvest was the highest when inoculated with *Bradyrhizobium* (68.13 cm). Kumari and Nair (2003) observed that the extent of plant growth were more in blackgram and greengram where *Bradyrhizobium* inoculation was done.

Table 4.11. Effect of bradyrhizobial inoculant on yield contributing characters of blackgram

Inoculant	Plant height (cm)	Pod length (cm)	Pods plant^{-1}	Seeds pod^{-1}	100-seed weight (g)
Non-inoculated	37.6b	3.47b	5.94b	4.70b	5.34
Inoculated	40.5a	3.96a	8.70a	5.06a	5.62
SE (±)	0.57	0.12	0.27	0.11	NS

In a column, means followed by different letter are significantly differed at 5% level by DMRT
NS = Not significant

4.12.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* on plant height was insignificant (Table 4.12). Inoculation when applied with phosphorus @ 30 kg ha^{-1} gave the highest plant height (43.5 cm) which was superior to all other treatments. The lowest plant height (37.1 cm) was observed with the treatment combination of P_0 x U.

Table 4.12. Interaction effect of phosphorus and bradyrhizobial inoculant on yield contributing characters of blackgram

Phosphorus x Inoculant	Plant height (cm)	Pod length (cm)	Pods plant^{-1}	Seeds pod^{-1}	100-seed weight (g)
P_0 x U	37.1	3.43	5.52	4.45	5.01
P_0 x I	38.3	3.68	7.65	4.98	5.30
P_{10} x U	38.0	3.46	5.78	4.65	5.13
P_{10} x I	38.6	3.82	7.86	5.00	5.46
P_{20} x U	36.8	3.66	6.33	4.73	5.76
P_{20} x I	42.2	4.53	10.55	5.33	6.28
P_{30} x U	39.1	3.58	6.05	5.08	5.68
P_{30} x I	43.5	3.98	8.66	5.15	5.88
P_{40} x U	37.3	3.20	6.03	4.60	5.13
P_{40} x I	40.0	3.81	8.80	4.85	5.20
SE (±)	NS	NS	NS	NS	NS
CV (%)	6.5	13.9	16.4	9.8	8.3

NS = Not significant
U: Non-inoculated, I: Inoculated

4.13 Pod length

4.13.1 Effect of phosphorus

Effect of phosphorus on pod length was insignificant (Table 4.10). The highest pod length (4.10 cm) was recorded with P_{20} level which was superior to all other treatments. The treatment P_{40} produced the lowest pod length (3.51 cm). Similar results were observed by Tomar (1998).

4.13.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculant significantly increased pod length (Table 4.11). Inoculated plants produced higher pod length (3.96 cm) than non-inoculated plants (3.47 cm).

4.13.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* on pod length was insignificant (Table 4.12). *Bradyrhizobium* inoculant when applied with phosphorus @ 20 kg ha^{-1} gave the highest pod length (4.53 cm) which was superior to all other treatments. The lowest pod length (3.20 cm) was recorded with P_{40} x U treatment combination. Srinivas and Shaik (2002) studied that the interactions effects between N and P were not significant for pod length.

4.14 Pods $plant^{-1}$

4.14.1 Effect of phosphorus

Application of phosphorus on pod number was highly significant (Table 4.10). The highest pod number (8.44 $plant^{-1}$) was recorded due to the application of P @ 20 kg ha^{-1} which was statistically similar with P_{30} and P_{40} and the lowest was recorded in P_0 level. Ali *et al.* (1995) reported that phosphorus increased pods in blackgram. Similar result were observed by Tomar (1998), Maqsood *et al.* (2001), Kumar *et al.* (2000) and Patel and Thakur (2003).

4.14.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculant showed significant difference in pod number (Table 4.11). Inoculated plants produced higher pod number (8.70 plant^{-1}) whereas uninoculated plants produced comparatively less pod number (5.94 plant^{-1}). Malik *et al.* (2002) studied that seed inoculation with *Rhizobium* application resulted in the highest number of pods plant^{-1} (22.47). Asraf *et al.* (2003) found that seed inoculation resulted in the highest number of pods plant^{-1}.

4.14.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* on pod number was insignificant (Table 4.12). The treatment combination P_{20} x I produced the highest pod number (10.55 plant^{-1}) and the treatment combination P_0 x U produced the lowest pod number (5.52 plant^{-1}). Srinivas and Shaik (2002) studied that the interactions effects between N and P were not significant for the number pods plant^{-1}.

4.15 Seeds pod^{-1}

4.15.1 Effect of phosphorus

Effect of phosphorus on seed pod^{-1} was insignificant (Table 4.10). The highest seed per pod (5.11) was recorded with P_{30} level which was the best to all other treatments. The lowest seed number (4.71 pod^{-1}) was observed in the P_0 level. The results corroborated with the findings of Tomar (1998), Maqsood *et al.* (2001) and Kumar *et al.* (2000).

4.15.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculant significantly increased seed number per pod (Table 4.11). Inoculated plants produced higher seed number (5.06 pod^{-1}) whereas non-inoculated plants produced lower seed number (4.70 pod^{-1}). Srinivas and Shaik (2002) studied that seed inoculation with *Rhizobium* culture increased number of seeds per pod relative to the control.

4.15.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* on seed number pod^{-1} was insignificant (Table 4.12). *Bradyrhizobium* inoculant when applied with phosphorus @ 20 kg ha^{-1} gave the highest seed (5.33 pod^{-1}) which was superior to all other treatments. The lowest seed (4.45 pod^{-1}) was recorded with P_0 x U treatment combination.

4.16 100-seed weight

4.16.1 Effect of phosphorus

Phosphorus increased weight of 100-seed significantly (Table 4.10) at 5% level of probability. Application of phosphorus @ 20 kg ha^{-1} gave the highest weight of 100-seed (6.02 g) which was statistically similar with P_{30} level. The lowest weight of 100-seed (5.16 g) was recorded with P_0 level which was statistically similar with P_{10} and P_{40} level. Ali *et al.* (1995) reported that phosphorus application increased 100-seed weight in blackgram. The results corroborated with the findings of Rao and Rao (1996), Mahmud *et al.* (1997), Maqsood *et al.* (2001), Kumar *et al.* (2000) and Patel and Thakur (2003).

4.16.2 Effect of *Bradyrhizobium* inoculant

The effect of *Bradyrhizobium* inoculant on 100-seed weight was insignificant (Table 4.11). Higher weight of 100-seed (5.62 g) was observed in inoculated plants and lower weight of 100-seed (5.34 g) was observed in non-inoculated plants. Malik *et al.* (2002) studied that seed inoculation with *Rhizobium* application resulted in the highest 1000-seed weight (42.27g). Srinivas and Shaik (2002) studied that seed inoculation with *Rhizobium* culture increased 1000-seed weight relative to the control.

4.16.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

Interaction effect of phosphorus x *Bradyrhizobium* on 100-seed weight was insignificant (Table 4.12). Inoculation with phosphorus @ 20 kg ha^{-1} gave the highest weight of 100-seed (6.28 g) which was superior to all other treatments. The lowest weight of 100-seed (5.01 g) was recorded with P_0 x U treatment combination.

4.17 N content in seed

4.17.1 Effect of phosphorus

It was observed that N content in seed did not differ significantly by phosphorus application (Table 4.13). The highest N content in seed (3.24%) was recorded with P_{20} level and the lowest (3.21%) was obtained by P_0 and P_{40} level. The results corroborated findings of Bhalu *et al.* (1995), Trivedi (1996), Rao and Rao (1996), Trivedi *et al.* (1997b) and Tomar *et al.* (2001).

Table 4.13. Effect of phosphorus on the content and uptake of N, P and protein yield in blackgram seed

P level (kg ha^{-1})	N content in seed (%)	N uptake by seed (mg plant^{-1})	P content in seed (%)	P uptake by seed (mg plant^{-1})	Protein content in seed (%)	Protein yield by seed (mg plant^{-1})
P_0	3.21	55.0c	0.28c	4.81c	20.1	344d
P_{10}	3.22	61.8c	0.30b	5.75b	20.1	387c
P_{20}	3.24	83.4a	0.32b	8.14a	20.3	520a
P_{30}	3.22	70.9b	0.35a	7.63a	20.1	442b
P_{40}	3.21	56.7c	0.34a	6.25b	20.0	369c
SE (±)	NS	2.20	0.006	0.21	NS	12.1

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT
NS = Not significant

4.17.2 Effect of *Bradyrhizobium* inoculant

Effect of *Bradryrhizobium* inoculant on N content in seed was significant (Table 4.14). Inoculated plants produced higher N content in seed (3.28 %) whereas non-inoculated plants produced lower N content in seed (3.16 %). Reddy and Mallaiah (2001) found that *Rhizobium* inoculation increased nitrogen content of fresh seeds in the inoculated plants, while that in non-inoculated controls was only 2.72%. They also reported that the plants inoculated with the AH isolate showed better nodulation and nitrogen content compared to the plants inoculated with the VM isolate. Chatterjee and Bhattacharjee (2002) noted that plants inoculated with *Bradyrhizobium* strains showed increased N content.

Table 4.14. Effect of bradyrhizobial inoculant on the protein content and uptake of N, P and protein yield in blackgram seed

Inoculant	N content in seed (%)	N uptake by seed (mg plant^{-1})	P content in seed (%)	P uptake by seed (mg plant^{-1})	Protein content in seed (%)	Protein yield by seed (mg plant^{-1})
Non-inoculated	3.16b	57.3b	0.30b	5.52b	19.7b	358b
Inoculated	3.28a	73.8a	0.33a	7.50a	20.5a	467a
SE (±)	0.006	1.40	0.003	0.13	0.04	7.6

In a column, means followed by different letters are significantly differed at 5% level by DMRT

4.17.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* inoculant of N content in seed was found statistically insignificant (Table 4.15). The highest N content in seed (3.31 %) was recorded with P_{20} x I treatment combination and the lowest N content in seed (3.13 %) was obtained by P_{10} x I treatment combination. Tomar *et al.* (2003) observed that P application and inoculation treatments increased N content in blackgram. Tanwar *et al.* (2003b) found that the N contents increased with increasing P rate up to 80 kg ha^{-1} with inoculation in blackgram.

Table 4.15. Interaction effect of phosphorus and bradyrhizobial inoculant on the protein content and uptake of N, P and protein yield in blackgram

Phosphorus x Inoculant	N content in seed (%)	N uptake by seed (mg plant^{-1})	P content in seed (%)	P uptake by seed (mg plant^{-1})	Protein content in seed (%)	Protein yield by seed (mg plant^{-1})
P_0 x U	3.15	49.5f	0.27	4.24f	19.7	309f
P_0 x I	3.27	60.5cde	0.29	5.37de	20.4	378de
P_{10} x U	3.15	55.3ef	0.29	5.07ef	19.7	346ef
P_{10} x I	3.13	68.3c	0.31	6.44c	20.5	428c
P_{20} x U	3.17	66.0cd	0.30	6.23c	19.8	412cd
P_{20} x I	3.31	100.8a	0.33	10.05a	20.7	629a
P_{30} x U	3.16	59.0de	0.33	6.16cd	19.8	368de
P_{30} x I	3.27	82.7b	0.36	9.10b	20.4	517b
P_{40} x U	3.16	56.6ef	0.33	5.93cd	19.8	357ef
P_{40} x I	3.25	56.8ef	0.35	6.57c	20.3	381cde
SE (±)	NS	3.12	NS	0.29	NS	17.1
CV (%)	0.9	9.5	5.4	8.9	0.8	8.3

In a column, means followed by a common letter are not significantly differed at 5% level by DMRT
NS = Not significant
U: Non-inoculated, I: Inoculated

4.18 N uptake by seed

4.18.1 Effect of phosphorus

Effect of phosphorus on N uptake by seed was highly significant (Table 4.13 and Figure 4.7). Application of phosphorus @ 20 kg ha^{-1} gave the highest N uptake by seed (83.4 mg plant^{-1}) which was significantly different from all other treatments. The treatments P_0 gave the lowest amount of N uptake by seed (55.0 mg plant^{-1}). The results are in line with Bhalu *et al.* (1995), Singh *et al.* (2002) and Poonkodi (2004).

4.18.2 Effect of *Bradyrhizobium* inoculant

Inoculation increased the amount of N uptake by seed which became statistically significant (Table 4.14 and Figure 4.8). Inoculated seeds uptook higher amount of N (73.8 mg plant^{-1}) whereas non-inoculated plants uptook lower amount of N (57.3 mg plant^{-1}). Srivastav and Poi (2000) found that inoculation with NK-4 into blackgram resulted in the highest nitrogen uptake.

4.18.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

Interaction effect of phosphorus and *Bradyrhizobium* on the amount of N uptake by seed was highly significant (Table 4.15). The highest amount of N uptake by seed (100.8 mg plant^{-1}) was recorded with *Bradyrhizobium* inoculant when applied with P @ 20 kg ha^{-1} which was significantly different from and superior to all other treatments. The lowest amount of N uptake by seed (49.5 mg plant^{-1}) was recorded with P_0 x U level. Tomar *et al.* (2003) observed that P application and inoculation treatments in blackgram increased N uptake by wheat significantly. Tanwar *et al.* (2003b) found that the N uptake increased with increasing P rate up to 80 kg ha^{-1} with inoculation in blackgram.

4.19 P content in seed

4.19.1 Effect of phosphorus

The effect of phosphorus on P content in seed (%) was significant (Table 4.13). Application of phosphorus @ 30 kg ha^{-1} gave the highest P content in seed (0.35 %) which was similar with P_{40} level but differed with other P levels. The lowest amount of P content in seed (0.28%) was recorded with P_0 level. Trivedi *et al.* (1996) reported that P content in seed increased with rate of P application. Similar results were observed by Rao and Rao (1996), Trivedi *et al.* (1997b), Thiyageshwari and Perumal (1999), Tomar *et al.* (2001) and Singh and Singh (2004).

4.19.2 Effect of *Bradyrhizobium* inoculant

Effect of *Bradryrhizobium* inoculant on P content in seed (%) was significant. Inoculated plants produced higher P content in seed (0.33%) than non-inoculated plants (0.30%).

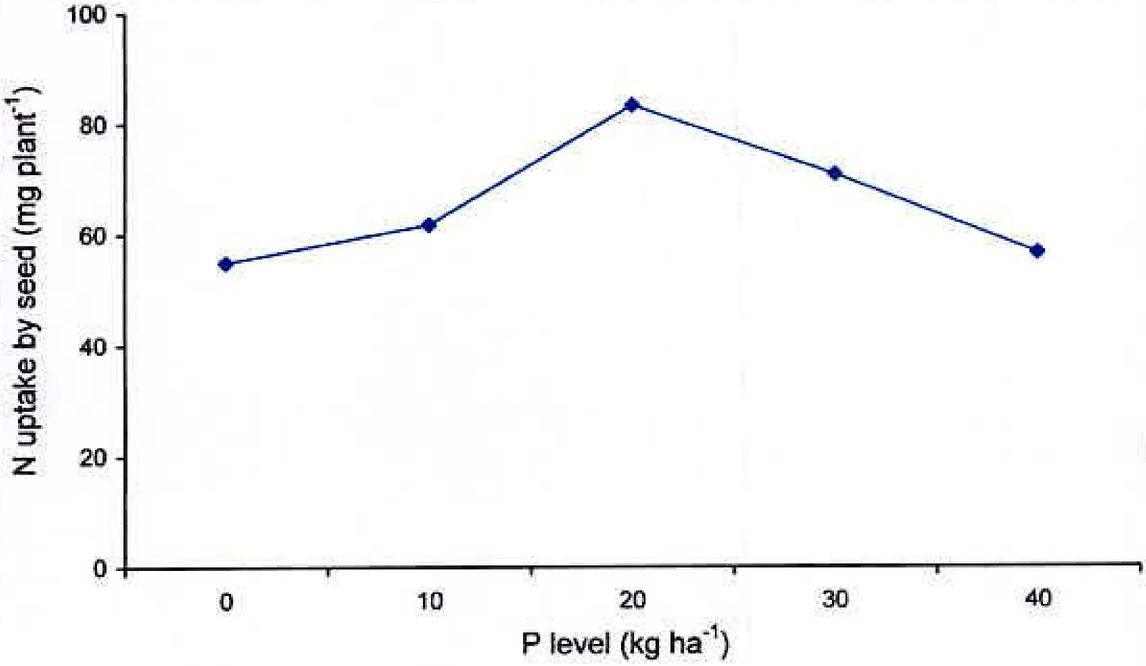

Fig. 4.7. Effect of different levels of phosphorus on N uptake by seed in blackgram

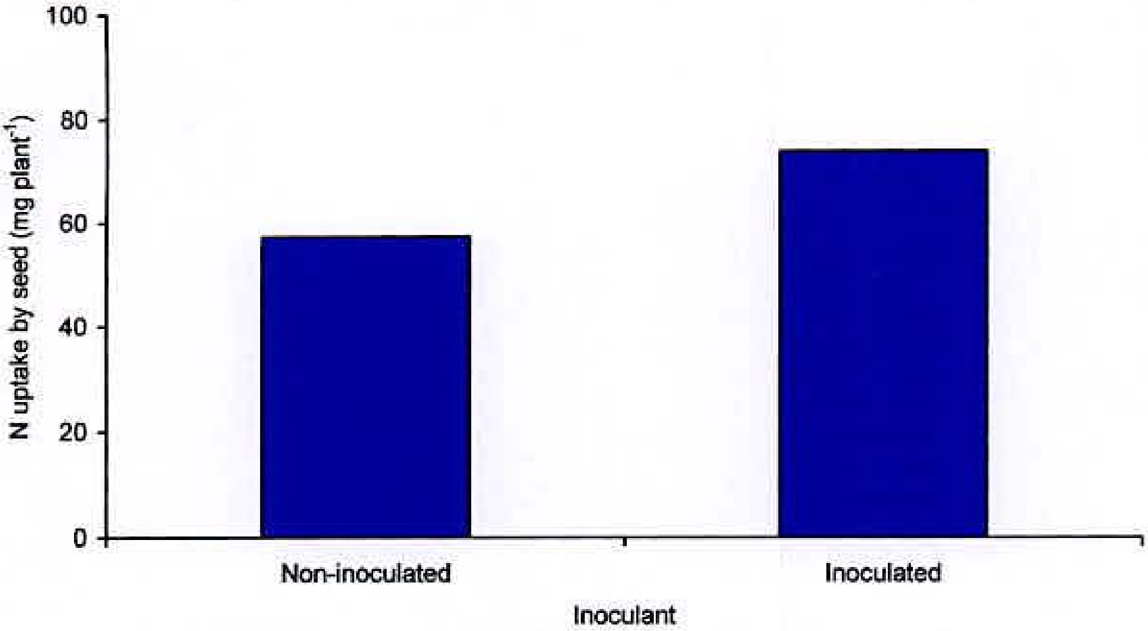

Fig. 4.8. Effect of bradyrhizobial inoculant on N uptake by seed in blackgram

4.19.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* inoculant on P content in seed was found statistically insignificant (Table 4.15). The highest P content in seed (0.36 %) was recorded with P_{30} x I treatment and the lowest P content in seed (0.27%) was obtained by P_0 x U treatment. Tomar *et al.* (2003) observed that P application and inoculation treatments increased P content in blackgram. Tanwar *et al.* (2003b) found that the P contents increased with increasing P rate up to 80 kg ha^{-1} with inoculation in blackgram.

4.20 P uptake by seed

4.20.1 Effect of phosphorus

Effect of phosphorus on P uptake by seed was highly significant (Table 4.13 and Figure 4.9). Application of phosphorus @ 20 kg ha^{-1} gave the highest P uptake by seed (8.14 mg $plant^{-1}$) which was statistically similar with P_{30} level. The treatments P_0 gave the lowest amount of P uptake by seed (4.81 mg $plant^{-1}$). Triyageshwari and Perumal (1999) reported that phosphorus uptake was higher where phosphorus was applied. Bhalu *et al.* (1995), Singh *et al.* (2002) and Poonkodi (2004) reported the similar results.

4.20.2 Effect of *Bradyrhizobium* inoculant

Bradyrhizobium inoculation increased the amount of P uptake by seed significantly (Table 4.14 and Figure 4.10). Inoculated plants absorbed higher amount of P by seed (7.50 mg $plant^{-1}$) than non-inoculated plant's seed of P (5.52 mg $plant^{-1}$).

4.20.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

Interaction effect on the amount of P uptake by seed was highly significant (Table 4.15). The highest amount of P absorbed by seed (10.05 mg $plant^{-1}$) was recorded with *Bradyrhizobium* inoculant with P @ 20 kg ha^{-1} which was significantly different from all other treatments. The lowest amount of P uptake by seed (4.24 mg $plant^{-1}$) was recorded with P_0 x U level. Tomar *et al.* (2003) observed that P application and inoculation treatments in blackgram increased P uptake by wheat significantly. Tanwar *et al.* (2003b) found that the P uptake increased with increasing P rate up to 80 kg ha^{-1} with inoculation in blackgram.

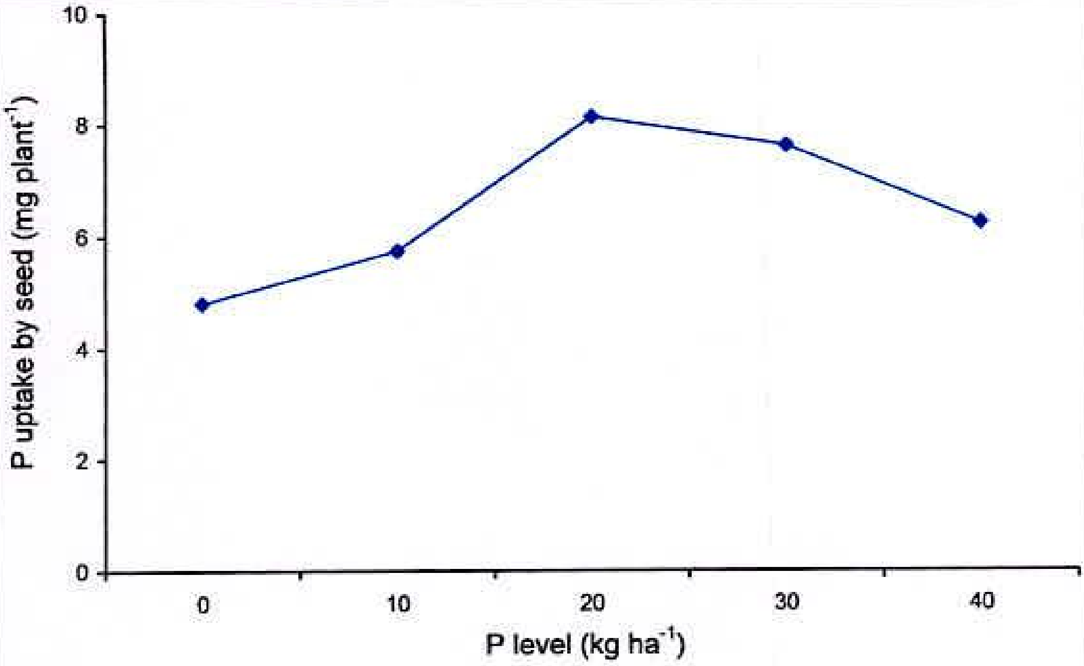

Fig. 4.9. Effect of different levels of phosphorus on P uptake by seed in blackgram

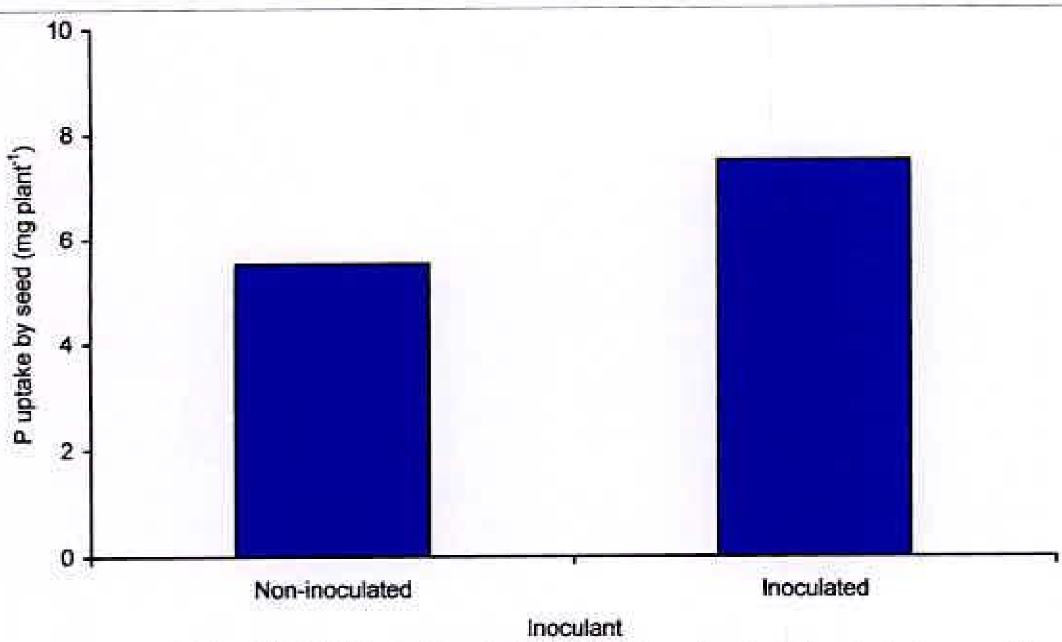

Fig. 4.10. Effect of bradyrhizobial inoculant on P uptake by seed in blackgram

4.21 Protein content in seed

4.21.1 Effect of phosphorus

It was observed that protein content in seed did not differ significantly by phosphorus application (Table 4.13). The highest protein content in seed (20.3%) was recorded with P_{20} level and the lowest (20.0%) with P_{40} level. Bhalu *et al.* (1995) reported that seed protein content increased with increasing N and P rates. Similar results were observed by Ali *et al.* (1995), Trivedi (1996) and Singh (2004).

4.21.2 Effect of *Bradyrhizobium* inoculant

Effect of *Bradryrhizobium* inoculant on protein content in seed (%) was significant (Table 4.14). Inoculated plants produced higher protein content of seed (20.5%) whereas non-inoculated plants produced lower protein content of seed (19.7%). Sharma *et al.* (1999) reported no significant effects in protein content by *Rhizobium* inoculation but slight improvement was observed over control.

4.21.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

The interaction effect of phosphorus x *Bradyrhizobium* inoculant on protein content in seed was found statistically insignificant (Table 4.15). The highest protein content in seed (20.7%) was recorded with P_{20} x I treatment and the lowest protein content in seed (19.7%) was obtained by P_0 x U and P_{10} x U treatments.

4.22 Protein yield by seed

4.22.1 Effect of phosphorus

Effect of phosphorus on protein yield by seed was highly significant (Table 4.13 and Figure 4.11). Application of phosphorus @ 20 kg ha^{-1} gave the highest protein yield in seed (520 mg plant^{-1}) which was significantly different from all other treatments. The treatments P_0 gave the lowest amount of protein yield by seed (344 mg plant^{-1}).

4.22.2 Effect of *Bradyrhizobium* inoculant

Inoculation increased the amount of protein yield by seed which became statistically significant (Table 4.14 and Figure 4.12). Inoculated plants produced higher amount of protein yield by seed (467 mg plant^{-1}) whereas non-inoculated plants produced lower amount of protein yield by seed (358 mg plant^{-1}).

4.22.3 Interaction effect of phosphorus and *Bradyrhizobium* inoculant

Interaction effect on the amount of protein yield by seed was highly significant at 5% level of probability (Table 4.15). The highest amount of protein yield by seed (629 mg plant) was recorded with *Bradyrhizobium* inoculant with P @ 20 kg ha^{-1} which was statistically different from all other treatments. The treatment P_0 x U produced the lowest amount of protein yield by seed (309 mg plant^{-1}).

4.23 Correlation

Correlation matrix among the plant characters of blackgram has been shown in Table 4.16. Most of the plant characters were strongly correlated among themselves. In the present study, nodule number had positive and significant correlation with nodule weight, root weight, shoot weight, root length, shoot length, seed yield, stover yield, N and P uptake and protein yield. Nodule weight had also positive and significant correlation with root weight, shoot weight, root length, shoot length, seed yield, stover yield, N, P uptake and protein yield. Root weight had positive and significant correlation with root length, shoot length, seed yield, stover yield, N, P uptake and protein yield. Shoot weight had positive and significant correlation with shoot length, seed yield, N uptake and protein yield. Root length had positive and significant correlation with shoot length, seed yield, N, P uptake and protein yield. Shoot length had positive and significant correlation with seed yield, N, P uptake and protein yield. Seed yield had positive and significant correlation with stover yield, N, P uptake and protein yield. Stover yield had positive and significant correlation with N, P uptake and protein yield. N uptake had positive and significant correlation with P uptake and protein yield. P uptake had positive and significant correlation with protein yield. Sarker *et al.* (2002) reported that correlation coefficient studies showed high correlation between seed yield and root weight, shoot length and shoot fresh weight. These results confirmed the findings of Khanam (2002). She observed positive and significant correlation of nodule number with nodule weight, root weight, and shoot weight of inoculated chickpea and soybean. Solaiman (1999) found positive correlation among mungbean growth, N uptake and yield parameters.

Table 4.16. Correlation matrix among different plant characters of blackgram (n=40)

Characters	Correlation coefficient (r value)									
	Nodule weight	Root weight	Shoot weight	Root length	Shoot length	Seed yield	Stover yield	N uptake	P uptake	Protein yield
Nodule number	0.849**	0.653**	0.609**	0.563**	0.627**	0.842**	0.383*	0.859**	0.826**	0.874*
Nodule weight		0.741**	0.570**	0.589**	0.756**	0.886**	0.475**	0.878**	0.882**	0.908**
Root weight			0.324^{NS}	0.536**	0.459*	0.656**	0.369*	0.635**	0.797**	0.677**
Shoot weight				0.338^{NS}	0.474**	0.457*	0.366^{NS}	0.488**	0.349^{NS}	0.480**
Root length					0.516**	0.491**	0.299^{NS}	0.545**	0.592**	0.554**
Shoot length						0.600**	0.390^{NS}	0.649**	0.654**	0.668**
Seed yield							0.418*	0.960**	0.911**	0.973**
Stover yield								0.446*	0.430*	0.454*
N uptake									0.927**	0.985**
P uptake										0.952**

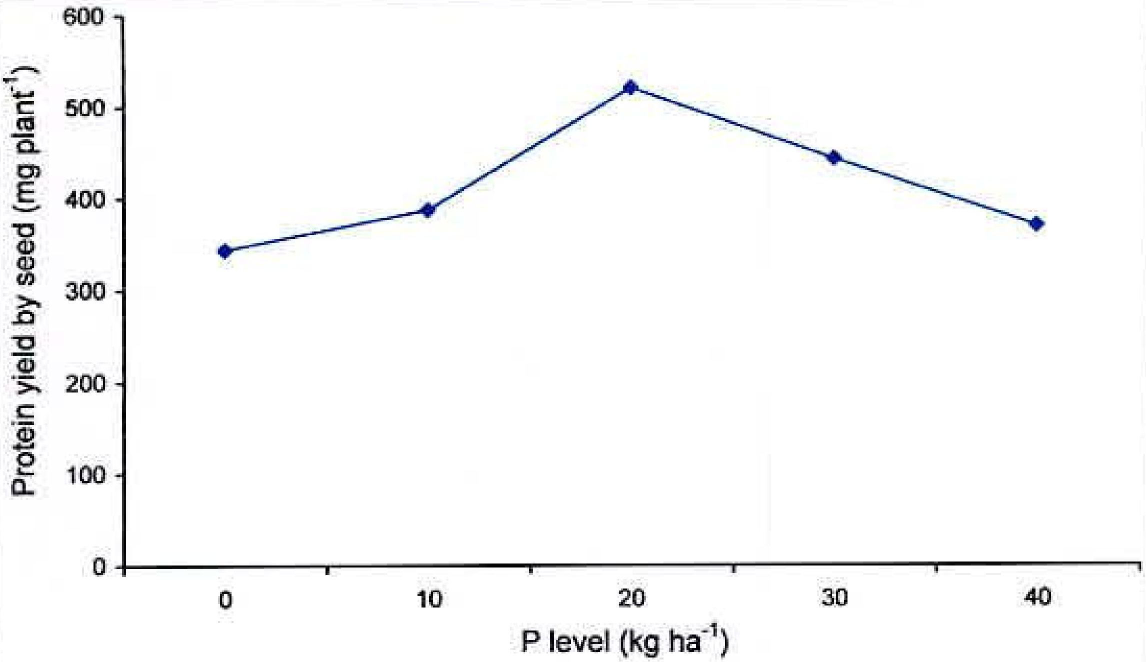

Fig. 4.11. Effect of different levels of phosphorus on protein yield by seed in blackgram

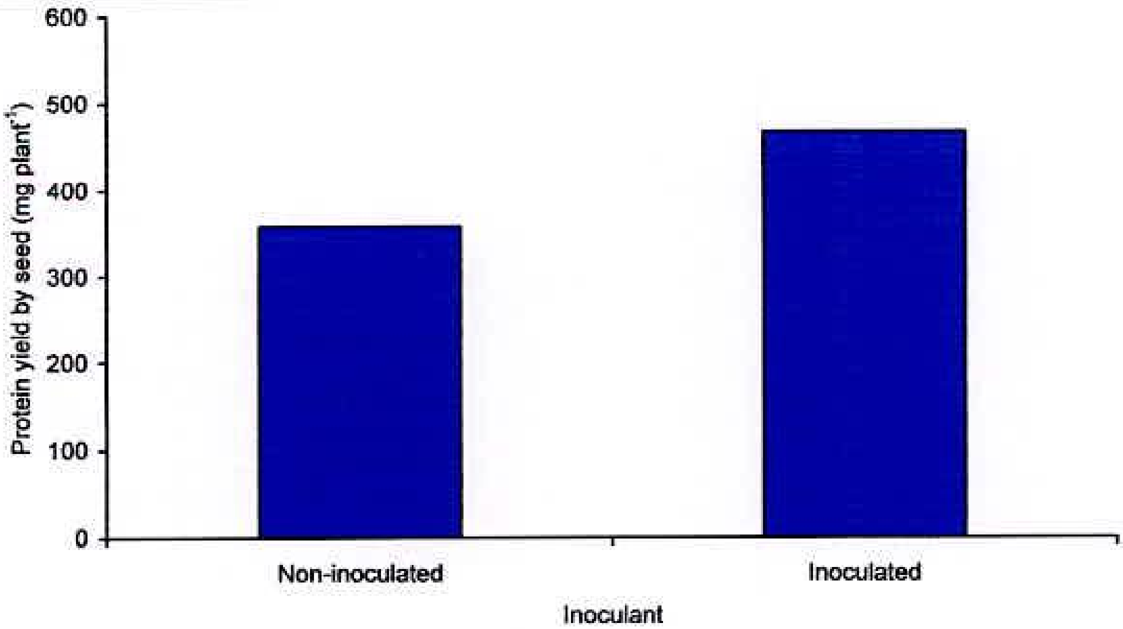

Fig. 4.12. Effect of bradyrhizobial inoculant on protein yield by seed in blackgram

Chapter 5

Summary, Conclusion and Recommendation

CHAPTER V

SUMMARY, CONCLUSION AND RECOMMENDATION

A pot culture experiment was conducted at the net house and laboratory of Soil Science Division in Bangladesh Agricultural Research Institute (BARI), Joyebpur, Gazipur during September to December 2007 to evaluate the performance of phosphorus levels and *Bradyrhizobium* on growth, nodulation, yield, nitrogen, phosphorus content and uptake, and other yield contributing characters and to determine the relationship between different parameters as affected by inoculation and phosphorus application.

There were 10 treatment combinations with two *Bradyrhizobium* inoculation treatments namely non-inoculated and inoculated and five levels of phosphorus @ 0, 10, 20, 30 and 40 kg ha^{-1}. There were 40 pots in the experiment arranged in factorial randomized complete block design. The earthen pot was filled with 10 kg of sieved soil. All chemical fertilizers except triple super phosphate (TSP) were applied as basal dose and as recommended levels at the time of pot filling. Blackgram seeds (BARI Mash-1) with inoculated and non-inoculated were sown on 5^{th} September, 2007, taking 20 seeds in each pot following dibbling method. Necessary water was added to the pots at a regular interval of 7 days until crop maturity to maintain proper moisture content.

Data on nodulation and dry matter production were recorded at two stages of growth viz. 35 and 50 days after sowing and for seed yield, stover yield, at the time of plant harvest. The crop was harvested at maturity and seed stover yield were recorded at 14% moisture content.

The seed was chemically analyzed for N, P content and uptake and protein content and protein yield. All the data were statistically analyzed by F-test and the differences between treatments means were adjudged by Duncan's Multiple Range Test (DMRT).

An abridged account of the result obtained is summarized below:

Significant influences of phosphorus levels were observed on nodulation, root length, yields (seed and stover), nutrient content and uptake by the seed. The highest total nodule number (16.00 and 20.00 plant^{-1}), nodule weight (19.25 and 72.50 mg plant^{-1}), shoot weight (1.55 and 4.58 g plant^{-1}), root length (7.63 and 8.50 cm) were obtained from P_{20} levels at both 35 and 50 DAS, respectively. The highest root weight (0.10 g plant^{-1} at 35 DAS) was observed in P_{20} level but in case of 50 DAS, the highest (0.39 g palnt^{-1}) was in P_{30} level. The highest shoot length was obtained form P_{30} level at 35 DAS, but incase of 50 DAS, it was obtained from P_{20} level. The treatment P_0 produced the lowest nodulation, root weight, root length, shoot length at both 35 and 50 DAS. But, the lowest shoot weight was observed in P_{40} at both 35 and 50 DAS.

The treatment P_{20} produced the highest pod yield (3.31 g plant^{-1}), seed yield (2.64 g plant^{-1}), stover yield (3.33 g plant^{-1}), pod length (4.10 cm) and 100-seed weight (6.02 g). Higher number of pods (8.44 plant^{-1}) was also recorded in P_{20} level. The lowest pod, seed yield, stover yield and 100-seed weight were recorded in P_0 level. Lower plant height pods plant^{-1}, seed pod^{-1} were also recorded in P_0 level.

Application of *Bradyrhizobium* inoculant produced significant effect on various crop characters. The highest total nodule number (13.30 and 21.17 plant^{-1}) and nodule weight (18.69 and 64.30 mg palnt^{-1}) were recorded in *Bradyrhizobium* inoculated pots at 35 and 50 DAS, respectively. Seed inoculation significantly increased seed, pod and stover yields of blackgram. *Bradyrhizobium* inoculation also significantly increased root weight, shoot weight, root length and shoot length, pods plant^{-1}, seed pod^{-1}, plant height and pod length. *Bradyrhizobium* inoculation also significantly increased nutrient content and uptake, and protein content and protein yield.

The interaction effect of phosphorus and *Bradyrhizobium* was significant on total nodule number (at 35 DAS) and nodule weight (at 50 DAS). Root weight (at 50 DAS) and shoot weight (at 35 DAS), pod yield, seed yield and N, P uptake and P content and protein yield.

The highest total number of nodules (17.00 and 27.75 palnt^{-1} at 35 and 50 DAS, respectively) and the highest shoot weight (1.79 and 4.91 g plant^{-1} at 35 and 50 DAS, respectively) was obtained from P_{20} x I treatment. The highest root and shoot length were also found in P_{20} x I level. The highest pod yield, seed yield and stover yield (4.04, 3.04 and 3.37 g plant^{-1}, respectively) were also obtained from P_{20} x I level. The highest 100 seed weight (6.28 gm) and protein yield (628.75 mg plant^{-1}) were also found in P_{20} x I level.

From the above investigation, it may be concluded that winter blackgram (BARI Mash-1) production in Agro-ecological Zone (AEZ-28) of Bangladesh will be increased in an expected level by using 20 kg P ha^{-1} chemical fertilizer along with effective culture of *Bradyrhizobium* inocula as biofertilizer. This technology not only improves soil organic matter content but also maintains soil health and keeps soil and environment free from pollution.

5.1 RECOMMENDATION AND SUGGESTION FOR FUTURE RESEARCH

1. Considering the trend of soil fertility decline, the use of bradyrhizobial inoculant and 20 kg phosphorus (P) per ha^{-1} should be used for cultivation of blackgram.

2. Instead of applying nitrogenous fertilizers for blackgram production, bio-fertilizer (bradyrhizobial inoculant) should be used. Because nitrogenous fertilizer is now a days a costly chemical fertilizer in Bangladesh. So, bradyrhizobial inoculant should be used in different pulses like blackgram for higher production of pulses to meet up the protein requirement of our sweet motherland, Bangladesh.

3. However, the effectivity of bradyrhizobial strain with various levels of P needs to be investigated in diverse soil and management condition.

Chapter 6
References

REFERENCES

Abd-Alla, M.H. 1994. Phosphorus and the utilization of organic phosphorus by *Rhizobium leguminosarum* biovar viceae. Lett. Appl. Microbiol 18: 294-296.

Afzal, M.A., Bakr, M.A. and Raham, M.L. 1999. Blackgram cultivation in Bangladesh. Lentil, Blackgram and Mungbean Development Pilot Project, Pulses Res. Station, BARI, Gazipur. 19: 64.

Agarwal, S.K., Behl, N.K. and Moolani, M.K. 1976. Response of summer mung to levels of phosphorus and irrigation under different dates of planting. Indian J. Agron. 21(3): 290-291.

Ali, C.K., Khan, T.R. and Javaid, M.A. 1995. Effect of fertilizer on growth, yield and protein content of mash (*Vigna mungo* L.). Fertiliser News. 40(2): 27, 29, 31.

Ashraf, M., Mueen-Ud-Din, M. and Warrick, N.H. 2003. Production efficiency of mungbean (*Vigna radiata* L.) as affected by seed inoculation and NPK application. Inter. J. Agric. Biol. 5(2): 179-180.

Balachandar, D., Nagarajan, P. and Gunasekaran, S. 2003. Effects of micronutrients on nodulation and yield of blackgram in acid soil conditions. Legume Res. 26(2): 153-154.

Bali, A.S., Sing, K.N., Shah, M.H. and Khandey, B.A. 1991. Effect of nitrogen and phosphorus fertilizer on yield and plant characters of mungbean (*Vigna radiata* L. Wilczek) under late sown condition of Kashmir valley. Fertilizer News, India. 36(7): 59-61.

BARC. 2005. Fertilizer Recommendation Guide. Bangladesh Agricultural Research Council. Farmgate, New Airport Road, Dhaka-1215.

BBS. 2006. Statistical Yearbook of Bangladesh. 26th Edit. Bangladesh Bureau of Statistics. Planning Division, Ministry of Planning, Govt. of the Peoples Republic of Bangladesh, Dhaka, Bangladesh. p. 143.

Bhalu, V.B., Sadaria, S.G., Kaneria, B.B. and Khanpara, V.D. 1995. Effect of nitrogen, phosphorus and *Rhizobium* inoculation on yield and quality, N and P uptake and economics of blackgram (*Phaseolus mungo*). Indian J. Agron. 40(2): 316-318.

Bhattacharyya, J. and Pal, A.Q. 2001. Effect of *Rhizobium* inoculation, phosphorus and molybdenum on the growth of summer greengram (*Vigna radiata* L. Wilczek). Indian Agric. 39(2): 173-179.

Bhuiyan, M.A.H., Mian, M.H. and Islam, M.S. 2006. Nodulation in five mungbean varieties with or without *Bradyrhizobium* inoculation. Bangladesh J. Microbiol. 23(1): 43-48.

Bhuiyan, M.A.H., Mian, M.H. and Islam, M.S. 2007b. Effect of *Bradyrhizobium* inoculation on dry matter production and nitrogen uptake in mungbean. Bangladesh J. Agric. and Environ. 3(2): 7-16.

Bhuiyan, M.A.H., Mian, M.H., Islam, M.S. and Islam, M.R. 2005. Detecting time of nodule initiation in mungbean. Bangladesh J. Microbiol. 22(1): 23-28.

Bhuiyan, M.A.H., Mian, M.H., Islam, M.S. and Islam, M.R. 2007a. Effects of inoculation with *Bradyrhizobium* on nodulation pattern, nodule senescence and size distribution in mungbean. Bangladesh J. Agril. Res. 32(2): 269-282.

Black, C.A. 1965. Methods of Soil Analysis, Part I. Agron. Monograph. 9. ASA Madison. Wisconsin. USA.

Borah, N. Borah, U.K., Goswami, J. and Deka, N. 2000. Effect of phosphorus and lime on soil acidity, forms of aluminium and yield of blackgram (*Vigna mungo* (L.) Hepper). Annals Biol. Ludhiana. 16(1): 3-6.

Bremner, J.M. and Mulvaney, C.S. 1982. Total Nitrogen *In*. Methods of Soil Analysis, Ed. Page, AL., Miller, R.H. and Keeney, D.R. 1982. Amer. Soc. Agron. Agron. Inc. Madison, Wisconsin. USA. pp. 595-622.

Chanda, M.C., Satter, M.A., Solaiman, A.R.M. and Podder, A.K. 1991. Effect of *Rhizobium* inoculation on mungbean varieties as affected by chemical fertilizers. Inter. Bot. Conf., 10-12 January 1991. Dhaka, Bangladesh. p. 9.

Chatterjee, A. and Bhattacharjee, P. 2002. Influence of combined inoculation and *Bradyrhizobium* and phosphobacteria on mungbeans in field. J. Mycol. Res. 40(2): 201-203.

Chaudhary, H.P. and Das, S.K. 1996. Effect of P, S and Mo application on yield of rainfed blackgram and their residual effect on safflower and soil and water conservation in an eroded soil. J. Indian Soc. Soil Sci. 44(4): 741-745.

Chowdhury, M.M.U., Ullah, M.H. and Mahmmud, Z.U. 2000. Dry matter production in mungbean (*Vigna radiata* (L) Wilczek) as influenced by *Bradyrhizobium* inoculation and phosphorus application. Legume Res. 23(1): 15-20.

Das, P.K., Sahoo, P.N. and Jena, M.K. 1997. Effect of VA-mycorrhiza and *Bradyrhizobium* inoculation on nutrient uptake in mungbean (*Vigna radiata*). Environ. Ecol. 15(4): 830-833.

Das, P.K., Sethi, A.K., Jena, M.K. and Patra, R.K. 1999. Effect of P sources and dual inoculation of VA-mycorrhiza and *Rhizobium* on dry matter yield and nutrient uptake by greengram (*Vigna radiata* L.). J. Indian Soc. Soil Sci. 47(3): 466-470.

Dey, K.B. 1988. Phosphate solubilizing organisms in improving fertility status. *In:* Biofertilizers Potentialities and Problems. Sen S.P Palit, P. Ed. Calcutta. Plant Physiology Forum, Naya Prokash. pp. 237.

Evans, J., Fettell, N.A., Coventry, D.R., O'Connor G.E., Walsgott, D.N., Mahoney, J. and Armstrong, E.L. 1991. Wheat response after temperate crop legumes in south easrtern Australia. Australian J. Agric. Res. 42: 31-43.

Fox, R.L., Olsen, R.A. and Rhoades, H.F. 1964. Evaluating the sulphur status of soil by plant and soil test. Soil Sci. Soc. Am. Proc. 28: 243-246.

Freed, R.D. 1992. MSTAT-C. Crop and Soil Science Department, Michigon State University, USA.

Ghosh, G. and Poi, S.C. 1988. Response of *Rhizobium*, phosphate solubilizing bacteria and microhizal organisms on some legume crops. Environ. Ecol. 16: 607-610.

Gunjkar, M.U., Dhoble, M.V., Dahiphale, V.V. and Jadhav, A.S. 1999. Response of blackgram (var. TAU-1) to nitrogen and phosphorus fertilization. J. Maharashtra Agric. Univ. 24(2): 211.

Hafeez, F.Y., Aslam, Z. and Malik, K.A. 1988. Effect of salinity and inoculation on growth, nitrogen fixation and nutrient uptake of *Vigna radiata* (L.) Wilczek. Plant and Soil 106: 3-8.

Jackson, M.L. 1973. Soil Chemical Analysis. Prentice Hall of India Pvt. Ltd. New Delhi, India.

Jayakumar, M., Eyini, M. and Velmurugan, R. 1997. Effect of seed *Rhizobium*-pelleting treatment and addition of coir-pith compost to soil on growth and nodulation of fluoride-water-irrigated blackgram (*Phaseolus mungo*). Indian J. Agric. Sci. 67(12): 601-603.

Kavathiya, Y.A. and Pandey, R.N. 2000. Interaction studies on *Meloidogyne javanica*, *Rhizobium* species and Macrophomina phaseolina in mungbean. J. Mycol. Plant Path.. 30(1): 91-93.

Keeney, D.R. and Nelson, D.W. 1982. Nitrogen inorganic Forms. *In*: Methods of Soil Analysis. Part 2. 2nd Ed. Page, A.L. Miller, R.H. and Keeney, D.R. 1982. Amer. Soc. Agron. Inc. Madison, Wisconsin., USA. pp. 643-693.

Khanam, D. 2002. Biodiversity of arbuscular mycorrhizal fungi in agricultural crops and their interaction with *Rhizobium* on chickpea (*Cicer arietinum* L.). Ph.D. Thesis. Bangabandhu Sheikh Mujibur Rahman Agric. Univ., Gazipur. pp. 1-226.

Knudsen, D., Petterson, G.A. and Pratt, P.F. 1982. Lithium, Sodium and Potassium, *In*. Methods of Soil Analysis, Part 2, 2nd Ed., Page. A.L., Miller. R.H. and Keeney, D.R. 1982. Amer Soc. Agron. Inc. Madi. Wis., USA. pp. 225-245.

Kumar, K., Verma, A.K. and Srivastava, G.P. 2000. Yield attributing character and grain yield of urdbean (*Vigna mungo* L. Hepper) as influenced by levels of phosphate application. J. Res. Birsa Agric. Univ. 12(2): 233-234.

Kumar, L.S.S., Agarwala, A.C., Arakeri, H.R. and Kamath, M.O. 1963. Pulses Agriculture in India. Asian Publishing House, New Delhi. Vol. 2, Crop. 38.

Kumari, K.S.M. and Nair, S.K. 2003. Use of acid tolerant strains of *Bradyrhizobium* sp. for blackgram and greengram. Legume Res. 26(1): 60-62.

Mahmud, M., Chiezey, U.F., Ahmed, M.K. and Rufai, I. 1997. Effects of different levels of phosphorus fertilizer and intra-row spacing on the grain yield, growth and yield components of black gram (*Vigna mungo* (L) Hepper). Discovery and Innovation. 9(1/2): 47-51.

Maldal, A.B. and Ray, R. 1999. Effect of *Bradyrhizobium* inoculation and nitrogenous fertilizer on the performance of mung. J. Interacademicia, 3(3-4): 259-262.

Malik, M.A., Hussain, S., Warraich, E.A., Habib, A and Ullah, S. 2002. Effect of seed inoculation and phosphorus application on growth, seed yield and quality of mungbean (*Vigna radiata* L.) cv. NM-98). Inter. J. Agric. Biol. 4(4): 515-516.

Manivannan, V., Thanunathan, K., Imayavaramban, V. and Ramanathan, N. 2003. Growth and growth analysis of rice-fallow blackgram as influenced by foliar application of nutrients with and without *Rhizobium* seed inoculation. Legume Res. 26(4): 296-299.

Maqsood, M., Mahmood-ul-Hassan, Hussain, M.I. and Mahmood, M.T. 2001. Effect of different levels of phosphorus on agronomic traits of two mashbean genotypes (*Vigna mungo* L.). Pakistan J. Agric. Sci. 38(1/2): 81-83.

Mathan, K.K., Francis, H.J. and Ramanathan, S.P. 1996. Response of blackgram (*Phaseolus mungo*) to fertilization and *Rhizobium* inoculation. Indian J. Agron. 41(1): 74-77.

Meenakumari, K.S. and Nair, S.K. 2001. Influence of soil chemical properties on root nodulation by *Bradyrhizobium* sp. in cowpea, blackgram and greengram. J. Trop. Agric. 39(1): 62-64.

Mishra, C.M. 1993. Response of blackgram (*Phaseolus mungo*) varieties to levels of phosphorus under rainfed conditions. Indian J. Agron. 38(3): 489-490.

Nagarajan, P. and Balachandar, D. 2001. Influence of *Rhizobium* and organic amendments on nodulation and grain yield of blackgram and greengram in acid soil. Madras Agric. J. 88(10/12): 703-705.

Nath, A.K. and Borach, D.K. 1983. A study on the release of native and applied fixed phosphate as affected by pH and moisture regime. Indian J. Agric. Chem. 16: 247-251.

Nelson, D.W. and L.E. Sommers. 1982. Total carbon, Organic carbon and Organic matter. *In*: Methods of Soil Analysis. Part. 2. 2nd Ed. Page, A.L., Miller, R.H. and Keeny, D.R. (Eds.). Amer. Soc. Agron. Madison, Wisconsin, USA. pp. 539-579.

Oikeh, S.O., Chude, V.O., Carsky, R.J., Weber, G.K. and Horst, W.J. 1998. Legume rotation in the moist tropical savana: managing soil nitrogen dynamics and cereal yields in farmers' fields. Expt. Agric. 34: 73-83.

Olsen, S.R. and Sommers, L.E. 1982. Phosphorus, *In*: Methods of Soil Analysis. Part II. 2nd Ed. Page, A.L. Miller, R.H. and Keeney, D.R. 1982. Amer. Soc. Agron. Inc. Madison, Wisconsin, USA. pp. 403-430.

Page, A.L., Miller, R.H. and Keeney, D.R. 1982. Methods of Soil Analysis. Part 2. 2nd Ed. Amer. Soc. Agron., Inc., Medison, Wisconsin, USA.

Parveen, S., Khan, M.S. and Zaidi, A. 2002. Effect of rhizospheric microorganisms on growth and yield of greengram (*Phaseolus radiatus*). Indian J. Agril. Sci. 72(7): 421-423.

Patel, S.R. and Thakur, D.S. 2003. Response of blackgram (*Phaseolus mungo*) to levels of phosphorus and phosphate solubilizing bacteria. Annals Agric. Res. 24(4): 819-823.

Patra, D.K. and Bhattacharyya, P. 1997. Influence of root nodule bacterium on nitrogen fixation and yield of mungbean. J. Mycopath. Res. 35(1): 47-49.

Poonkodi, P. 2004. Phosphorus use efficiency in black gram with press mud. Adv. in Plant Sci. 17(1): 239-241.

Ramamoorthy, K., Balasubramanian, A. and Arokiaraj, A. 1997. Response of rainfed blackgram (*Phaseolus mungo*) to phosphorus and sulphur nutrition in red lateritic soils. Indian J. Agron. 42(1): 191-193.

Rao, V.U. and Rao, A.S. 1996. Response of blackgram (*Phaseolus mungo*) and greengram (*P. radiatus*) to inoculation with vesicular-arbuscular mycorrhizal fungus under different sources of phosphorus fertilizer. Indian J. Agric. Sci. 66(10): 613-616.

Reddy, A.S. and Mallaiah, K.V. 2001. Studies on the effect of *Rhizobium* inoculation on blackgram. Bioinoculants for sustainable agriculture and forestry. Proc. Nat. Symp. held on February 16-18, 2001. pp. 93-98.

Reddy, B.P. and Swamy, S.N. 2000. Effect of farmyard manure, phosphate solubilising bacteria and phosphorus on yield and economic of blackgram (*Phaseolus mungo*). Indian J. Agric. Sci.70(10): 694-696.

Reeves, T.J., Ellington, A. and Brooke, H.D. 1984. Effects of lupin-wheat rotations on soil fertility crop disease and crop yields. Australian J. Exp. Agric. Animal Husbandry. 24: 595-600.

Rhoades, J.D. 1982. Cation Exchange Capacity. *In*: Methods of Soil Analysis. Part-2, 2nd Ed. (Page, A.L., Miller, R.H. and Keeney, D.R. eds.). 1982. Amer. Soc. Agron., Inc., Madison, Wisconsin, USA. pp. 149-157.

Richardson, A.E. 1994. Soil microorganisms and phosphorus availability. *In:* Soil Biota, Management in Sustainable Farming Systems, Pankhurst, C.E. Double, B.M. Grupta V.V.S.R. and Grace, P.R. Eds. Melbourne, Australia: CSIRO. pp. 50-62.

Roasales, C.M., Rivera, F.G., Hautea, R.A. and Del Rosario, E. 1998. Field evaluation of N_2 fixation by mungbean in the Philippines and residual effects on maize. *In*: Improving yield and nitrogen fixation of grain legumes in the tropics and sub-tropic of Asia. pp. 65-75. (IAEA TECDOC-1027; Vienna).

Sampathkumar, G. and Ganeshkumar, A. 2003. Effect of AM fungi and *Rhizobium* on growth and nutrition of *Vigna mungo* L., and *Vigna unguiculata* L. Mycorrhiza News. 14(4): 15-18.

Sarkar, H. K., Pal, A.K and Baisya, R. 2002. Effect of *Bradyrhizobium* inoculation on black gram [*Vigna mungo* (L.) Hepper] with special reference to root and nodule parameters. J. Interacademicia. 6(3): 260-265.

Satter, M.A. and Ahmed, S.U. 1995. Response of mungbean (*Vigna radiata* L. Wilczek) to inoculation with *Bradyrhizobium* as affected by phosphorus levels. Proc. Inter. Cong. Com. IV. 419-423.

Shaikh, M.A.Q. 1977. Grain legumes in Bangladesh. Induced Nutritions for the Improvement of grain legume production in South Eash Asia. IAEA-TDCDOC-203. Inter. Atomic Energy Agency, Vienna. pp. 61-77.

Shanmugam, V., Renukadevi, P., Senthil, N., Raguchander, T. and Samiyappan, R. 2001. Interaction of *Pseudomonas fluorescens* with *Rhizobium* in the management of blackgram root rot incited by *Macrophomina phaseolina*. Plant Disease Res. 16(1): 23-29.

Sharma, P. and Khurana, A.S. 1997. Effect of single and multistrain *Rhizobium* inoculants on biological nitrogen fixation in summer mungbean, *Vigna radiata* (L.) Wilczek. Res. and Dev. Reporter. 14(1-2): 8-11.

Sharma, S., Sharma, C.R. and Upadhayay, R.G. 1999. Response of various strains of *Rhizobium* and nitrogen on symbiotic and physiological parameters, biochemical constituents and yield of blackgram (*Vigna mungo* L. Hepper). Legume Res. 22(4): 233-236.

Sharma, S., Upadhyay, R.G. and Sharma, C.R. 2000. Effect of *Rhizobium* inoculation and nitrogen on growth, dry matter accumulation and yield of blackgram (*Vigna mungo* (L.) Hepper). Legume Res. 23(1): 64-66.

Sharma, S.C., Tiwari, H.O. and Tiwari, R.J. 1995. Effect of nitrogen, phosphorus and sulphur on the micronutrient content of blackgram. Crop Res. Hisar. 9(1): 54-58.

Sheikh, J., Akhter, A., Khan, N.A. and Inam, A. 2003. Effect of *Rhizobium* and NPK on growth, physiomorphological traits and yield of wastewater grown blackgram. Indian J. Pulses Res. 16(1): 25-30.

Shrivastava, U.K. and Rajput, R.L. 2000. Effect of PSB on seed yield of black gram (*Phaseolus mungo*). Bhartiya Krishi Anusandhan Patrika. 15(1/2): 39-41.

Shukla, S.K. and Dixit, R.S. 1996a. Nutrient and plant population management in summer greengram (*Phaseolus radiatus*). Indian J. Agron. 41(1): 78-83.

Shukla, S.K. and Dixit, R.S. 1996b. Effect of *Rhizobium* inoculation, plant population and phosphorus on growth and yield of summer greengram (*Phaseolus radiatus*). Indian J. Agron. 41(4): 611-615.

Singh, R.K.K., Athokpam, H.S., Singh, L.N. and Chongtham, N. 2002. Effect on nitrogen, phosphorus and potassium on the growth, yield and nutrient uptake of blackgram (*Vigna mungo*, L). J. Agric. Sci. Soc. North East India. 15(2): 175-177.

Singh, S. and Kapoor, K.K. 1994. Solubilization of insoluble phosphate by bacteria isolated from different sources, Environ. Ecol. 12: 51-55.

Singh, Y.P. 2004. Role of sulphur and phosphorus in blackgram production. Fertilizer News. 49(2): 33-36.

Singh, Y.P. and Singh, R. 2004. Interaction effect of sulphur and phosphorus on growth and nutrient content of black gram (*Phaseolus mungo* L.). J. Indian Soc. Soil Sci. 52(3): 266-269.

Singha, D.D. and Sarma, A. 2001. Response of blackgram (*Phaseolus mungo*) to phosphorus fertilization and *Rhizobium* inoculation in the hill soils of Assam. Annals Agril. Res. 22(1): 151-153.

Solaiman, A.R.M. 1999. Response of mungbean to *Bradyrhizobium* sp. (*Vigna*) inoculation with and without phosphorus and potassium fertilization. Bangladesh J. Sci. Res. 17(2): 125-132.

Srinivas, M. and Shaik, M. 2002. Effect of *Rhizobium* inoculation and different levels of nitrogen and phosphorus on yield and economics of greengram (*Vigna radiata* L. Wilczek.). Crop Res. 24(3): 463-466.

Sriramachandrasekharan, M.V. and Vaiyapuri, V. 2003. Influence of granular insecticide-carbofuran in association with *Rhizobium* on nodulation, growth and yield of blackgram. J. Ecobiol. 15(2): 107-111.

Srivastav, A.K. and Poi, S.C. 2000. Response of greengram (*Vigna radiata*) and blackgram (*Vigna mungo*) to *Bradyrhizobium* as well as the residual effects of *Bradyrhizobium* strains on the same crops. Adv. Plant Sci. 13(2): 431-435.

Stevenson, F.J. and Cole, M.A. 1999. Cycles of Soil: Carbon, Nitrogen, Phosphorus, Sulphur, Micronutrients, 2nd Edn. Wiley, New York, USA. p. 427.

Tanwar, S.P.S., Sharma, G.L. and Chahar, M.S. 2002. Effects of phosphorus and biofertilizers on the growth and productivity of blackgram. Annals Agric. Res. 23(3): 491-493.

Tanwar, S.P.S., Sharma, G.L. and Chahar, M.S. 2003a. Economics of black gram (*Vigna mungo* (L.) Hepper) cultivation as influenced by phosphorus levels and biofertilizers. Legume Res. 26(2): 149-150.

Tanwar, S.P.S., Sharma, G.L. and Chahar, M.S. 2003b. Effect of phosphorus and biofertilizers on yield, nutrient content and uptake by blackgram (*Vigna mungo* (L.) Hepper). Legume Res. 26(1): 39-41.

Thakur, A.K. and Panwar, J.D.S. 1985. Effect of *Rhizobium*-VAM interactions on growth and yield in mungbean (*Vigna radiata* L. Wilczek) under field condition. Indian J. Plant Physiol. 38: 62-65.

Thakur, N.S. 1999. Effect of phosphorus, and sulphur levels with and without *Rhizobium* inoculation on growth and yield of blackgram (*Vigna mungo* L.). Adv. Agric. Res. in India. 12: 67-70.

Thiyageshwari, S. and Perumal, R. 2000. Changes in available phosphorus and grain yield of blackgram (*Vigna mungo*) under integrated nutrient management in Inceptisol. Agropedology 10(1): 40-43.

Tomar, A., Kumar, N., Pareek, R.P. and Chaube, A.K. 2001. Synergism among VA mycorrhiza, phosphate solubilizing bacteria and *Rhizobium* for symbiosis with blackgram (*Vigna mungo* L.) under field conditions. Pedosphere. 11(4): 327-332.

Tomar, A., Kumar, N., Pareek, R.P. and Chandra, R. 2003. Residual effect of blackgram inoculated *Rhizobium*, VAM and PSB on succeeding wheat crop. Indian J. Pulses Res. 16(2): 141-143.

Tomar, R.K.S. 1998. Effect of phosphate-solubilizing bacteria and farmyard manure on the yield of blackgram (*Phaseolus mungo*). Indian J. Agric. Sci. 68(2): 81-83.

Trivedi, S.K. 1996. Response of blackgram (*Phaseolus mungo*) to nitrogen, phosphorus and sulphur. Legume Res. 19(1): 7-9.

Trivedi, S.K., Singh, V. and Shinde, C.P. 1997a. Effect of nitrogen, phosphorus and sulphur on yield and chemical composition of blackgram (*Phaseolus mungo*). Gujarat Agric. Univ. Res. J. 22(2): 46-50.

Trivedi, S.K., Singh, V., Shinde, C.P. and Patil, R. 1997b. Effect of N, P and S on yield and nutrient uptake of blackgram (*Phaseolus mungo*). Bhartiya Krishi Anusandhan Patrika. 12(2): 90-94.

Vincent, J.M. 1970. Selective Indicator Media. A. Manual for the practical study of the root nodule bacteria. IBP Hand Book No. 15: 4.

Yadav, K.S. and Dadarwal, K.R. 1997. Phosphate solubilization and mobilization through soil microorganisms. *In:* Biotechnological Approaches in Soil Microorganisms for Sustainable Crop Production. Dadarwal, R.K. Ed. Scientific Publishers, Jodhpur, India. pp. 293-308.