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Responses of Pea Varieties to *Rhizobium* Inoculation: Nitrogenase Activity, Dry Matter Production and Nitrogen Uptake

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ABSTRACT : The responses of five varieties and three cultivars of pea (*Pisum sativum*) to *Rhizobium* inoculation on nodulation, growth, nitrogenase activity, dry matter production and N uptake were investigated. The pea varieties were IPSA Motorshuti-1, IPSA Motorshuti-2, IPSA Motorshuti-3, BARI Motorshuti-1, BARI Motorshuti-2 and the cultivars were 063, Local small and Local white. Fifty percent seeds of each pea variety/cultivar were inoculated with a mixture of *Rhizobium* inoculants at rate of 15 g/kg seed and the remaining fifty percent seeds were kept uninoculated. The plants inoculated with *Rhizobium* inoculant significantly increased nodulation, growth, nitrogenase activity, dry matter production and N uptake. Among the varieties/cultivars, BARI Motorshuti-1 performed best in almost all parameters including nitrogenase activity of root nodule bacteria of the crop. There were positive correlations among the number and dry weight of nodules (r=0.987, 0.909**), nitrogenase activity of root nodule bacteria (r=0.944**, 0.882**), dry weight of shoot (r=0.787**, 0.952**), N content (r=0.594**, 0.605**) and N uptake (r=0.784**, 0.922**) by shoot both at flowering and pod filling stages of the crop, respectively. It was concluded that BARI Motorshuti-1 in symbiotic association with *Rhizobium* inoculant performed best in recording nitrogenase activity, dry matter production and N uptake by pea.**

***Keywords*:** Pea, *Rhizobium sp. (Pisum)*, nodulation, nitrogenase activity, dry matter production, N uptake

Nitrogen is the most deficient nutrient element in Bangladesh soil limiting crop production. Urea fertilizer is generally used for enriching soil nitrogen. But the cost of urea fertilizer is high and it causes injury to soil health. Pea is one of the most important food legumes in Bangladesh. Pea like many other legumes is capable of fixing and utilizing atmospheric nitrogen through symbiotic relationship with *Rhizobium* bacteria at the root of the crop. It was reported that *Rhizobium* inoculation added 80 kg N/ha and average dry matter yield was increased by pea plants (Micanovic *et al.,* 1997) over the uninoculated control. Significant increase in biological nitrogen fixation and pod yield were obtained by *Rhizobium* inoculation of peas (Feng *et al.,* 1997). Biological nitrogen fixation and seed yield of soybean and chickpea were significantly increased due to inoculation (Solaiman, 1999a, 1999c). Results of a field trial showed that *Rhizobium* inoculation and different levels of nitrogen increased nodulation, growth and N uptake of lentil (Mahmud *et al.*, 1997).

Bangladesh Agricultural Research Institute and Bangabandhu Sheikh Mujibur Rahman Agricultural University of Bangladesh released a number of high yielding pea varieties. Some local cultivars are also available in this country. But their comparative performances in respect of nodulation, biological nitrogen fixation, dry matter production and N uptake in association with *Rhizobium* inoculant have not been studied. Selection of pea varieties with high nodulation and nitrogen fixing abilities is very important for obtaining maximum growth and yield of pea. Keeping these facts in mind the present investigation was therefore, carried out to evaluate the responses of a number of pea varieties/cultivars to *Rhizobium* inoculation on nodulation, growth, nitrogenase activity, dry matter production and N uptake by the crop and to select a variety having high potential in nitrogenase activity, dry matter production and N uptake by pea.

MATERIALS AND METHODS

The experiment was conducted in earthen pot (24 cm×27 cm) containing 12 kg of air-dried soil to study the responses of five high yielding varieties and three local cultivars of pea (*Pisum sativum*) to *Rhizobium* sp. (*Pisum*) inoculation on nodulation, growth, nitrogenase activity, dry matter production and N uptake. The soil was of sandy loam texture and contained 0.95% organic carbon, 12.74 (meq/100 g soil) CEC, 0.085% total N, 16 ppm available P and 0.389 (meq/100 g soil) exchangeable K and had a pH 6.5. Basal doses of phosphorous, potassium and molybdenum were applied to the soil at the rate of 60 kg P_2O_5, 45 kg K_2O and 1.5 kg Mo/ha in the form of triple superphosphate, muriate of potash and ammonium molybdate, respectively. No nitrogenous

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fertilizer was used in the experiment.

The five high yielding pea varieties, IPSA Motorshuti-1, IPSA Motorshuti-2, IPSA Motorshuti-3, BARI Motorshuti-1 and BARI Motorshuti-2, and the three local cultivars, 063, Local small and Local white were used as the test crop. Two peat-based *Rhizobium* inoculants one containing the local strains BAU 303 and BAU 349 and the other containing the local strains BAU 424 and BAU 444 were collected from Bangladesh Agricultural University, Mymensingh, Bangladesh and used for inoculation of seeds. The inoculants were mixed together to ensure that if one or more of the strains of the inoculants would fail to form nodules and fix nitrogen due to some adverse environmental conditions, the strain which would survive in soil would form nodules and continue fixing nitrogen. *Rhizobium* population of the inoculant containing the strains BAU 303 and BAU 349 was 1.86×10^8 cells/g of peat soil and that of BAU 424 and BAU 444 was 4.78×10^8 cells/g of peat soil. Detailed information on the strains of *Rhizobium* sp. is given in Table 1. Fifty percent seeds of each variety/cultivar were inoculated with the mixed culture of *Rhizobium* inoculants at the rate of 15 g/kg seed using 40% solution of gum Arabic as sticking agent and the remaining fifty percent seeds were kept uninoculated. Inoculated seeds were air-dried before sowing. There were sixteen treatment combinations comprising of eight pea varieties/cultivars and two levels of *Rhizobium* inoculation viz. uninoculated and inoculated. Uninoculated and inoculated treatments were designated by R_0 and R_1, respectively. The experiment was laid out in a completely randomized design (CRD) with three replications. Three plants were maintained in each pot. Plant samples were collected from the pot at flowering and pod filling stages of the crop. From each pot three plants were carefully uprooted so that no nodule was left in the soil. The roots were washed thoroughly with water. Nitrogenase activity of the plants was assessed by measuring acetylene reducing activity (ARA) in a gas chromatograph (Shimadzu, GC-

Table 1. Some characteristics of the strains of *Rhizobium* sp. used in this experiment.

Strains of *Rhizobium* sp.	Sources of collection	Effectiveness in N_2 fixation on pea
BAU 303	*Lens culinaris*	Effective
BAU 349	*Lens culinaris*	Effective
BAU 424	*Lathyrus sativas*	Effective
BAU 444	*Lathyrus sativas*	Effective

Table 2. Effects of variety and *Rhizobium* inoculation on number and dry weight of nodule per plant at flowering and pod filling stages of pea.

Treatments		Number of nodules/plant		Dry weight of nodules (mg/plant)	
		Flowering stage	Pod filling stage	Flowering stage	Pod filling stage
IPSA Motorshuti-1	R_0	18.33 e	51.33 i	34.00 hi	71.57 k
	R_1	50.00 c	70.67 fg	92.00 f	163.20 ef
IPSA Motorshuti-2	R_0	21.67 e	46.33 i	35.00 hi	74.32 k
	R_1	53.33 bc	76.33 ef	100.00 e	165.20 e
IPSA Motorshuti-3	R_0	20.00 e	73.67 ef	32.33 i	98.25 i
	R_1	70.33 a	108.70 b	124.00 ab	201.20 b
BARI Motorshuti-1	R_0	21.00 e	98.33 bc	40.00 h	120.20 h
	R_1	72.00 a	125.00 a	130.00 a	208.00 a
BARI Motorshuti-2	R_0	18.00 e	62.67 gh	30.00 i	72.14 k
	R_1	60.00 b	95.00 c	116.00 cd	178.10 c
063	R_0	9.33 f	54.67 hi	19.00 j	70.31 k
	R_1	55.00 bc	83.00 de	110.00 d	158.6 f
Local small	R_0	8.67 f	50.00 i	18.00 j	59.11 l
	R_1	40.00 d	70.67 fg	84.00 g	135.8 g
Local white	R_0	15.00 ef	62.67 gh	29.00 i	81.01 j
	R_1	58.00 b	89.33 cd	120.00 bc	172.1 d

Means in a column followed by some letter (s) are not significantly different at 5% level of DMRT

8A) fitted with a flame ionization detector and a stainless steel column (3 mm dia., 1.2 m length). The nodules from the roots of each plant were separately collected and counted. The shoot, root and nodule materials were first air-dried and then oven-dried at 65°C for 72 hours, then the weights of dried shoot, root and nodule were recorded. The oven-dried plant shoot material was ground in a grinding machine (Wiley Cutting Mill, Model 1029-B, Yoshida Seisakusho Co. Ltd. Japan). Total N content in the shoot material was determined by ashing the plant material using salicylic acid modified kjeldahl method following sulfuric acid digestion and then steam distillation and titration assay (Wilde *et al.,* 1979). Nitrogen uptake by shoot was calculated from the data on dry matter yield and nitrogen content in shoot material of the crop. The recorded data on various characters of the crop were statistically analyzed to find out the significance of variation resulting from the treatments. The differences between the treatment means were compared by Duncans Multiple Range Test (DMRT). Correlations between different characters were also calculated.

RESULTS AND DISCUSSION

Number of nodules per plant

BARI Motorshuti-1 in combination with *Rhizobium* inoculation produced the highest number of nodules at flowering (72.00/plant) and pod filling (125.00/plant) stages (Table 2). The effect of this treatment was superior to other treatments except IPSA Motorshuti-3×R_1 at flowering stage. The effects of BARI Motorshuti 2×R_1 and Local white×R_1 were statistically similar and ranked second position but their effects were identical to IPSA-Motorshuti-2×R_1 and 063×R_1. The lowest number of nodules in uninoculated and inoculated conditions was produced by the cultivar Local small at flowering stage. At pod filling stage, IPSA Motorshuti-3×R_1 ranked second position in nodule number in inoculated condition. The lowest number of nodules in uninoculated and inoculated conditions was produced by both IPSA Motorshuti-1 and Local small. At flowering and pod filling stages BARI Motorshuti-1×R_1 recorded 243% and 240% higher number of nodule over its uninoculated condition, respectively. It was observed that *Rhizobium* inoculated plants produced significantly higher number of nodules over uninoculated plants. Feng *et al.* (1997) conducted a number of pot and field experiments on many cultivars of peas and observed 100% nodule in pot experiments and above 90% in field experiment due to *Rhizobium* inoculation. Jha and Singh (1996) showed several nodulation traits of inoculated peas cv. Rachna and HUP-2. Between the inoculated cultivars, nodule number was more promising in cv. HUP-2 than Rachna. Almost similar results in nodulation of inoculated peas were reported by Zafran *et al.* (1996).

Dry weight of nodules per plant

All the varieties/cultivars of pea recorded higher dry weight of nodules due to *Rhizobium* inoculation compared to uninoculated plants (Table 2). Results show that at flowering (130 mg/plant) and pod filling (208 mg/plant) stages BARI Motorshuti-1 in association with *Rhizobium* inoculation produced maximum dry weight of nodules but its effect was statistically similar to IPSA Motorshuti-3 at flowering stage. Dry weight of nodule produced by Local white×R_1 ranked second position but its effect was statistically similar to IPSA Motorshuti-3×R_1 and BARI Motorshuti-2×R_1 at flowering stage. The lowest dry weight of nodule in inoculated condition was obtained by Local small at both the stages. In uninoculated condition the lowest dry weight of nodules was observed by Local small and 063 at flowering stage. At pod filling stage, the second highest dry weight of nodules was found in IPSA Motorshuti-3×R_1 which was superior to all other inoculated treatments except BARI Motorshuti-1×R_1. The lowest dry weight of nodule at pod filling stage was found by Local small in uninoculated condition. At flowering and pod filling stages BARI Motorshuti-1 recorded 225% and 73% higher nodule weight over its uninoculated condition, respectively. Similar result was also obtained by Vaishya and Dube (1988) in their experiment where chickpea seed was inoculated with four *Rhizobium* strains. Solaiman (1999a) recorded the highest dry weight of nodule in soybean receiving inoculant+1.5 Mo ha^{-1}. Dry weight of nodule was increased by 1100% in inoculated chickpea as compared to control (Alam *et al.,* 1999). In the present study there was a strong positive correlation between the number and dry weight of nodules per plant both at flowering (r=0.987**) and pod filling (r=0.909**) stages of the crop (Table 7).

Nitrogenase activity

Rhizobium inoculated plants irrespective of variety/cultivar recorded higher nitrogenase activity than the uninoculated plants. Among the treatments, BARI Motorshuti-1×R_1 scored the highest nitrogenase activity both at flowering (64.78 μ mole C_2H_4/plant/hr) and pod filling (43.84 μ mole C_2H_4/plant/hr) stages and its effect was statistically superior to other treatments except IPSA Motorshuti-3×R_1 at flowering stage (Table 3). BARI Motorshuti-1 recorded 197% and 322% higher nitrogenase activity over its uninoculated condition at flowering and pod filling stages, respectively. Under uninoculated condition the lowest nitrogenase activity was

Table 3. Effects of variety and *Rhizobium* inoculation on nitrogenase activity of pea at flowering and pod filling stages.

Treatments		Nitrogenase activity (μ mole C_2H_4/plant/hour)	
		Flowering stage	Pod filling stage
IPSA Motorshuti-1	R_0	9.31 i	4.45 i
	R_1	28.19 e	10.52 g
IPSA Motorshuti-2	R_0	17.08 g	7.43 h
	R_1	49.39 c	30.78 c
IPSA Motorshuti-3	R_0	17.93 fg	9.50 g
	R_1	61.66 ab	40.39 b
BARI Motorshuti-1	R_0	21.79 f	10.39 g
	R_1	64.78 a	43.84 a
BARI Motorshuti-2	R_0	21.31 f	9.24 g
	R_1	60.50 b	39.34 b
063	R_0	14.66 gh	4.54 i
	R_1	34.66 d	14.16 f
Local small	R_0	12.29 hi	4.50 i
	R_1	38.40 d	19.39 e
Local white	R_0	17.88 fg	7.82 h
	R_1	52.38 c	28.04 d

Means in a column followed by same letter (s) are not significantly different at 5% level of DMRT

Table 4. Effects of variety and *Rhizobium* inoculation on plant height and root length at flowering and pod filling stages of pea.

Treatments		Plant height (cm)		Root length (cm)	
		Flowering stage	Pod filling stage	Flowering stage	Pod filling stage
IPSA Motorshuti-1	R_0	30.85 g	36.53 h	16.58 cde	21.29 def
	R_1	43.95 de	48.20 e	22.00 b	26.67 bc
IPSA Motorshuti-2	R_0	32.84 g	35.79 h	10.10 h	14.27 h
	R_1	45.33 d	50.49 de	13.00 fgh	21.53 def
IPSA Motorshuti-3	R_0	40.67 ef	43.27 f	14.20 efg	19.10 efg
	R_1	57.50 bc	62.80 b	19.50 bc	24.27 bcd
BARI Motorshuti-1	R_0	45.95 d	51.38 de	22.50 b	27.73 b
	R_1	58.67 b	61.36 b	30.33 a	34.60 a
BARI Motorshuti-2	R_0	28.84 g	42.96 fg	18.02 cd	20.94 def
	R_1	64.00 a	67.50 a	28.70 a	32.67 a
063	R_0	28.53 g	34.39 hi	12.20 gh	18.41 fg
	R_1	48.33 d	52.85 cd	15.00 defg	23.33 cde
Local small	R_0	29.33 g	32.39 i	12.44 gh	15.53 gh
	R_1	40.33 ef	43.06 fg	16.25 cdef	21.51 def
Local white	R_0	38.83 f	39.79 g	13.07 fhg	14.08 h
	R_1	53.66 c	55.67 c	17.83 cd	25.00 bcd

Means in a column followed by same letter (s) are not significantly different at 5% level of DMRT.

observed in IPSA Motorshuti-1 but its effect was statistically similar to Local small at flowering stage. At pod filling stage, IPSA Motorshuti-1 x R_0 recorded the lowest nitrogenase activity which was statistically identical to 063 x R_0 and Local small x R_0. IPSA Motorshuti-1 x R_1 recorded the lowest nitrogenase activity among the inoculated treatments at both the stages. This result is in agreement with that of Rai (1988) who conducted experiment with chickpea. Eusuf Zai *et al.* (1999) found significantly higher nitrogenase activity due to inoculation of chickpea. They found 254% and 660% higher nitrogenase activity over its uninoculated control at preflowering and flowering stages, respectively. Similar results in soybean and chickpea were observed by Solaiman (1999a, 1999c). Table 7 shows that nitrogenase activity was positively correlated with the number of nodules (r=0.944**, r=0.882**), dry weight of nodules (r=0.928**, r=0.848**) and shoot (r=0.773**, r=0.868**) of the crop both at flowering and pod filling stages, respectively.

Plant height

Plants inoculated with *Rhizobium* inoculant showed higher plant height than the plants receiving no inoculation. Among the varieties, BARI Motorshuti-2×R_1 recorded the highest plant height both at flowering (64.00 cm) and pod filling (67.50 cm) stages (Table 4). The second highest plant height was recorded by BARI Motorshuti-1×R_1 at flowering stage but its effect was statistically similar to IPSA Motorshuti-3×R_1. Local white x R_1 and IPSA Motorshuti-3×R_1 also gave similar result in recording plant height at flowering stage. At pod filling stage, statistically similar plant height was obtained by BARI Motorshuti-1×R_1 and IPSA Motorshuti-3×R_1. Local white×R_1 and 063×R_1 recorded similar plant height but the effect of 063×R_1 was statistically identical to IPSA Motorshuti-2×R_1. At flowering stage, the lowest plant height in inoculated condition was found by Local small and IPSA Motorshuti-1 and in uninoculated condition it was found by 063 whose effect was however, statistically similar to IPSA Motorshuti-1, IPSA Motorshuti-2, BARI Motorshuti-2 and Local small. At pod filling stage, the lowest plant height in inoculated condition was observed by Local small and in uninoculated condition it was observed by Local small and 063. This result is in agreement with the findings of Alam *et al.* (1999) who conducted experiment with chickpea. Sekhon *et al.* (1984) found 55.8 cm plant height in inoculated soybean plant whereas it was 48.4 cm in uninoculated control.

Root length

The highest root length was recorded by BARI Motorshuti-1×R_1 both at flowering (30.33 cm) and pod filling (34.60 cm) stages of the crop but the effect of this treatment was statistically similar to BARI Motorshuti-2×R_1 both at flowering and pod filling stages (Table 4). The second highest root length at flowering stage was observed by IPSA Motorshuti-1 and IPSA Motorshuti-3 in association with inoculation but in pod filling stage it was obtained by IPSA Motorshuti-1×R_1, IPSA Motorshuti-3×R_1 and Local white×R_1. Both in inoculated and uninoculated conditions the lowest root length was found by IPSA Motorshuti-2 at flowering stage. The effect of the

Table 5. Effects of variety and *Rhizobium* inoculation on dry weight of shoot and root per plant at flowering and pod filling stages of pea.

Treatments		Dry weight of shoot (mg/plant)		Dry weight of root (mg/plant)	
		Flowering stage	Pod filling stage	Flowering stage	Pod filling stage
IPSA Motorshuti-1	R_0	1180 l	2180 l	200 fgh	373 ef
	R_1	1670 i	2690 k	300 bcd	448 bcd
IPSA Motorshuti-2	R_0	1630 j	2750 j	213 efgh	380 def
	R_1	2460 f	3320 h	283 bcde	430 cde
IPSA Motorshuti-3	R_0	3040 d	4100 e	160 ghi	210 h
	R_1	3770 b	5320 b	313 abc	450 bc
BARI Motorshuti -1	R_0	3050 d	4270 d	230 defg	290 g
	R_1	3950 a	5410 a	373 a	520 a
BARI Motorshuti-2	R_0	1450 k	1910 m	183 fgh	340 fg
	R_1	3640 bc	4050 ef	350 ab	510 ab
063	R_0	810 n	1330 o	160 jhi	347 fg
	R_1	2390 g	3640 g	257 cdef	400 cdef
Local small	R_0	500 o	1070 p	150 hi	347 fg
	R_1	1830 h	2930 i	277 cde	427 cde
Local white	R_0	1070 n	1420 n	103 i	137 i
	R_1	3430 c	4450 c	193 fgh	280 g

Means in a column followed by same letter (s) are not significantly different at 5% level of DMRT treatment was statistically similar to 063 and Local small in inoculated condition and that of 063, Local small and Local white in uninoculated condition. At pod filling stage, the lowest root length in inoculated condition was recorded by Local small but its effect was statistically similar to IPSA Motorshuti-2×R_1, 063×R_1, IPSA Motorshuti-3×R_1 and Local white ×R_1. The lowest root length in uninoculated condition was obtained by the Local white followed by IPSA Motorshuti-2×R_0 and local small×R_0. Root length of the plants both at flowering and pod filling stages increased due to *Rhizobium* inoculation compared to uninoculated plants. This result has the resemblance with the result of Alam *et al.* (1999).

Dry weight of shoot

Results reported in Table 5 show that the highest dry weight of shoot was found in BARI Motorshuti-1 in association with *Rhizobium* inoculation both at flowering (3950 mg/plant) and pod filling (5410 mg/plant) stages. The second highest dry weight of shoot was recorded by IPSA Motorshuti-3×R_1 at both the stages. BARI Motorshuti-1×R_1 recorded 30% and 27% higher dry weight of shoot over its uninoculated control at flowering and pod filling stages, respectively. IPSA Motorshuti-1×R_1 showed the lowest dry weight of shoot both at flowering and pod filling stages and in control condition it was found by local small. It was observed that *Rhizobium* inoculation significantly increased dry weight of shoot of all the varieties/cultivars of pea as compared to their uninoculated control. Micanovic *et al.* (1997) stated that average dry matter yield of pea increased by *Rhizobium* inoculation over uninoculated control. Mahmud *et al.* (1997) reported that dry weight of shoot in lentil significantly increased due to inoculation compared to control. Dry weight of shoot remarkably increased in inoculated mungbean over control as reported by Solaiman (1999b). In the present study the number of nodule per plant had a positive correlation with the dry weight of shoot at flowering (r=0.787**) and pod filling (r=0.952**) stages of the crop. Dry weight of nodules also showed similar relationship with the dry weight of shoot at flowering (r=0.757**) and pod filling (r=0.839**) stages of the crop (Table 7).

Dry weight of root

Both at flowering (373 mg/plant) and pod filling (520 mg/plant) stages, maximum dry weight of root was observed in BARI Motorshuti-1 in association with *Rhizobium* inoculation (Table 5). The effect of BARI Motorshuti-1×R_1 was however, statistically similar to BARI Motorshuti-2 ×R_1 and IPSA Motorshuti-3×R_1 at flowering stage and BARI Motorshuti-2×R_1 at pod filling stage of the crop. The second highest dry weight of root was produced by IPSA Motorshuti-1 x R_1 followed by IPSA Motorshuti-2×R_1, 063×R_1 and Local small×R_1 at flowering stage. The second highest dry weight of root at pod filling stage was observed by IPSA Motorshuti-3×R_1 which was statistically identical to IPSA Motorshuti-1×R_1, IPSA Motorshuti-2×R_1, 063×R_1 and Local small×R_1. The lowest dry weight of root in inoculated condition was recorded by Local white both at flowering and pod filling stages, but it was statistically similar to 063×R_1 at flowering stage. In uninoculated condition the lowest dry weight of root was found in Local white at both the stages but its effect was statistically similar to Local small at flowering stage. However, irrespective of variety/cultivar *Rhizobium* inoculation significantly increased dry weight of root compared to the plants receiving no inoculation. Ashgar *et al.* (1988) observed that *Rhizobium* inoculation had a significant positive influence on dry weight of root. Mahmud *et al.* (1997) found that dry weight of root significantly increased due to inoculation over control.

Nitrogen content in shoot

The shoots of pea plants accumulated significant amount of nitrogen due to *Rhizobium* inoculation compared to uninoculated plants. At flowering stage (Table 6), the highest nitrogen content (4.24%) was recorded in BARI Motorshuti-1×R_1, but the effect of the treatment was statistically similar to BARI Motorshuti-2×R_1 and IPSA Motorshuti-3×R_1. There was no significant difference of the effects of IPSA Motorshuti-3×R_1, BARI Motorshuti-2×R_1 and 063×R_1. The lowest N content in shoot at this stage was observed in Local white and IPSA motorshuti-1 in inoculated and uninoculated conditions, respectively. The effects of 063×R_1, Local white×R_1, IPSA Motorshuti-1×R_1, IPSA Motorshuti-2×R_1 and Local small×R_1 were statistically similar and ranked second position in recording N content in shoot at this stage. At pod filling stage, BARI-Motorshuti-1×R_1 also accumulated the highest amount of N (3.21%) in shoot which was statistically superior to other treatments except IPSA Motorshuti-3×R_1 and BARI Motorshiti-2×R_1. At this stage lower amount of N was accumulated in shoot of IPSA Motorshuti-1×R_1, local small×R_1 and 063×R_1. IPSA Motorshuti-1×R_0, 063×R_0 and local small×R_0 recorded the lowest amount of nitrogen in shoot. Similar result was reported in lentil by Mahmud *et al.* (1997). Maurya *et al.* (1987) explained that inoculation of seeds with *Rhizobium* strains increased nitrogen content in chickpea. Data presented in Table 7 shows that N content in shoot had a strong positive correlation with the number of nodules at flowering (r=0.594**) and pod filling (r=0.605**) stages of the crop. N content in shoot also showed strong positive correlation with the dry weight of

Table 6. Effects of variety and *Rhizobium* inoculation on N content in shoot and N uptake by shoot of pea at flowering and pod filling stages.

Treatments		N content in shoot (%)		N uptake by shoot (mg/plant)	
		Flowering stage	Pod filling stage	Flowering stage	Pod filling stage
IPSA Motorshuti-1	R_0	2.66 e	1.82 i	31 gh	40 i
	R_1	3.15 cde	2.24 gh	53 ef	60 g
IPSA Motorshuti-2	R_0	3.12 cde	2.16 gh	51 ef	59 g
	R_1	3.16 cde	2.69 cde	78 d	89 f
IPSA Motorshuti-3	R_0	2.81 de	2.70 cde	85 d	111 e
	R_1	3.94 ab	3.05 ab	148 b	162 b
BARI Motorshuti-1	R_0	3.54 bc	2.87 bcd	108 c	123 cd
	R_1	4.24 a	3.21 a	167 a	174 a
BARI Motorshuti-2	R_0	2.96 cde	2.45 efg	43 fg	47 h
	R_1	3.88 ab	2.95 abc	141 b	119 b
063	R_0	2.97 cde	2.11 hi	24 hi	28 j
	R_1	3.40 bcd	2.44 efg	81 d	88 f
Local small	R_0	2.87 de	2.08 hi	14 i	22 k
	R_1	3.27 cd	2.28 fgh	60 e	67 g
Local white	R_0	2.80 de	2.57 def	30 gh	36 i
	R_1	3.10 cde	2.89 bc	106 c	129 e

Means in a column followed by same letter(s) are not significantly different at 5% level of DMRT.

Table 7. Relationship between different characters of pea recorded at flowering and pod filling stages.

Factors	Flowering stage			Pod filling stage		
	Number of nodules	Dry weight of nodules	Dry weight of shoot	Number of nodules	Dry weight of nodules	Dry weight of shoot
Dry weight of nodules	0.987	–	–	0.909	–	–
Nitrogenase activity	0.944	0.928	0.773	0.882	0.848	0.868
Dry weight of shoot	0.787	0.757	–	0.952	0.839	–
N content in shoot	0.594	0.563	–	0.605	0.795	–
N uptake by shoot	0.784	0.734	–	0.922	0.919	–

n=48 r value: $P_{0.05}$= 0.325 $P_{0.01}$= 0.418

nodule at flowering (r=0.563**) and pod filling (r=0.795**) stages of the crop.

N uptake by shoot

Rhizobium inoculation significantly increased N uptake by shoot of all the varieties/cultivars of pea compared to uninoculated plants. Among the varieties, BARI Motorshuti-1 in combination with *Rhizobium* inoculation recorded the highest N uptake both at flowering (167 mg/plant) and pod filling (174 mg/plant) stages (Table 6). The effect of the treatment was statistically superior to other treatments. The effect of IPSA Motorshuti-3×R_1 ranked second position in recording N uptake by shoot at both the stages, but it was statistically similar to BARI Motorshuti-2×R_1. Lower N uptake by shoot was obtained with IPSA Motorshuti-1×R_1, Local small×R_1 and local small×R_0 both at flowering and pod filling stages. Bhuiya *et al.* (1983) observed higher N uptake by grasspea due to inoculation with different *Rhizobium* strains. Muniruzzaman and Khan (1990) carried out an experiment and found that inoculation had a great effect on N uptake. Khan *et al.* (1997) reported that N uptake by shoot in lentil significantly increased due to inoculation along with nitrogen fertilizer. There was a positive correlation between N uptake by shoot and the number of nodules at flowering (r=0.784**) and pod filling (r=0.922**) stages of the crop. Similar relationship between N uptake by shoot and dry weight of nodule was also observed at flowering (r= 0.734**) and pod filling (r=0.919**) stages (Table 7).

In conclusion we have shown that inoculation of pea with *Rhizobium* sp. increased nodulation and thereby increased nitrogenase activity of root nodule bacteria. As a result the crop fixed higher atmospheric nitrogen and thus inhanced growth, dry matter production, N content and N uptake by pea. Among the varieties/cultivars, BARI Motorshuti-1 performed best. However, the effects of BARI Motorshuti-2 and IPSA Motorshuti-3 were comparable to BARI Motorshuti-1 in some of the parameters of the crop studied.

REFERENCES

Alam, M.J., A.R.M. Solaiman., A.J.M.S. Karim, and M.T. Hossain. 1999. Potential of some *Rhizobium* strains on nodulation, nitrogen fixation, crop growth and dry matter production of chickpea. *Bangladesh J. Microbiol.* 16 : 107-114.

Asghar, A.B., B. Roider Khan, and J.H. Keating. 1988. Effects of inoculation and phosphate fertilizer on lentil under rainfed conditions in upland Baluctistan. *Lens Newsletter* 15 : 29-33.

Bhuiya, Z.H., M.R. Islam., A.B.M. Alam, and M.S. Hoque. 1983. Effect of different locally isolated strains of *Rhizobium leguminosarum* on Bangladesh. *J. Soil. Sci.* 19: 63-67.

Eusuf Zai, A.K.E., A.R.M. Solaiman, and J.U. Ahmed. 1999. Response of some chickpea varieties to *Rhizobium* inoculation in respect of nodulation, Biological nitrogen fixation and dry matter yield. *Bangladesh J. Microbiol.* 16 : 135-144.

Feng, Y., C. Pan., D. Wang., Y. Li., C. Wei, and Y. Feng. 1997. Isolation of nodule bacteria from *Pisum sativum* and the application of nitragine from the isolate. *J. Trop. Sub Trop. Bot.* 5 : 47-53.

Jha, S.K., and V.P. Singh. 1996. Correlation for effectiveness of *Rhizobium leguminosarum* in pea (*Pisum sativum*). *J. Appl. Biol.* 6 : 19-21.

Khan, M.S.H., A.R.M. Solaiman., M.S. Hoque, and M.M. Rahman. 1997. Response of chickpea to *Rhizobium* inoculation and NPK fertilization: Nodulation, dry matter production and N uptake. *Ann. Bangladesh Agric.* 7 : 21-26.

Mahmud, M.S., A.R.M. Solaiman, M.S. Hoque, and M.A. Hashem. 1997. Influence of *Rhizobium* inoculant and nitrogen on nodulation, growth and N uptake of lentil. *Ann. Bangladesh Agric.* 7 : 139-143.

Maurya, B.R., C.L. Sanorria, and P.C. Ram. 1987. Combined culture treatment enhanced nodulation, yield and quality of chickpea. *Seeds and Farms.* 13 : 25-33.

Micanovic, D., Z. Saric., V. Raicevic., S. Jevtic, and B. Lazic. 1997. Possibility of nitrogen fixation in *Pisum sativum* and *Triticum aestivum.* Proc. of the First Balkan Symposium on Vegetables and Potatoes, Belgrade, Yegoslavia, 4-7 June. Acta Hort. 2(462): 823-827.

Muniruzzaman, S., and S.I. Khan. 1990. Symbiotic effectiveness and nodulation potential of streptomycin and streptomycin resistant mutants of some indigenous *Rhizobium* isolates. *Bangladesh J. Microbiol.* 7 : 19-23.

Rai, R. 1988. Interaction response of *Glomus albidus* and *Cicer arietinum* strains on iron uptake and symbiotic nitrogen fixation in calcareous soil. *J. Plant Nutr.* 11 : 863-869.

Sekhon, H. S., J.N. Kaul., and K.K. Dhingra. 1984. Effect of *Rhizobium* inoculation, mulch, nitrogen, and phosphorous fertilizer on soybean (*Glycine. max.* L. Meril). *J. Agric.* 10 : 43-47.

Solaiman, A.R.M. 1999a. Effects of *Bradyrhizobium japonicum* inoculation and molybdenum on soybean. *Bangladesh J. Bot.* 28 : 181-183.

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

Solaiman, A.R.M. 1999c. Nitrogenase activity, yield and yield contributing characters of chickpea as influenced by *Rhizobium* inoculant in presence and absence of nitrogen. *Bangladesh J. Sci. & Tech.* 1 : 141-144.

Vaishya, U.K., and J.N. Dube. 1988. Interaction between *Rhizobium* strains and chickpea varieties. *Agric. Sci. Digest.* 8 : 153-156.

Wilde, S.A., R.B. Corey, and J.G. Iyer. 1979. Soil and Plant Analysis for Tree Culture. Soil Science, University of Wisconsin, G.K. Voigt, Yale School of Forestry. p.78.

Zafran, J., S. Redzepovic, and V. Gamulin. 1996. Characterization two strains of *Rhizobium leguminosarum* biover *Viciae* efficient in the nodulation of host plants. *Poljoprivredna Znanstvena Smotra.* 61(3-4) : 157-166.