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Effect of Inoculation of Screened *Rhizobial* Isolates on the Growth of Pea (*Pisum sativum* (*L.*))

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A B S T R A C T

The present study was undertaken to obtain the best host cultivar-*Rhizobial* strain combination in the soils of Hoshiarpur. A Pot experiment was conducted by sowing seeds of Pea (*Pisum sativum* (L.)) varAP-3 along with screened isolated strains of *Rhizobia* in soil samples. After 60 days of sowing various parameters such as number of nodules, fresh weight and dry weight of plants, length of plants, leghaemoglobin content, nitrogenase activity and percent nitrogen content were recorded. Maximum number of nodules was observed in SS8 (53) followed by strain SS12 (20). The maximum fresh weight (11.35g/root and 12.20g/shoot), dry weight (8.17g/root and 9.16g/shoot)and length (20.3 cm/root and 17.0 cm/shoot)were reported in plants inoculated with SS8 strain of *Rhizobia*. The leghaemoglobin content in nodules was found to be highest in plants inoculated with SS8 strain (8.96 mg/0.5 g nodules) followed by plants inoculated with SS12 strain of *Rhizobia* (6.36 mg/0.5 g nodules). The nitrogenase activity (0.65 µM C_2H_2 reduced/plant) was also observed to be highest in plants inoculated with SS8 strain of *Rhizobia*. The total nitrogen content was 3.22% in SS8 inoculated plant leaves while it was 2.10% in SS12 inoculated plant leaves.

Introduction

Pea (*Pisum sativum* (L.)) is an important pulse crop of Punjab and it contains considerable amounts of carbohydrates, vitamins along with digestible proteins. These leguminous plants have high nutritive and biomass producing values. The key element nitrogen comprises about 79% of total atmospheric gases and is responsible for enhancing crop production by converting atmospheric nitrogen into plant usable form.

All leguminous plants such as Pea, Alfalfa, Mungbean, Chickpea etc. contain *Rhizobium leguminosarum* strains in their nodules. These *Rhizobial* strains are able to fix approximately 360 Kg of N/ha/year in association with

different leguminous crops (Peoples *et al.*, 1995; Sibbal *et al.*, 2002; Sheng and Jing, 2003). Legume-*Rhizobium* symbiosis not only depends on the capability of introduced strains of *Rhizobia* that combat over native *Rhizobia* for nodulation to enhance its capacity but also is affected by the genetic characters of the host plant (Gresshoff *et al.*, 2014; Stephen *et al.*, 2019). The present study was undertaken with the objective to develop best Host-Rhizobial combinations.

Materials and Methods

Rhizobium leguminosarum strains were isolated from healthy nodules of PeavarAP-3 grown in soils of Hoshiarpur. Gram staining was done to ensure purity and freedom from Gram-positive bacteria. Seeds of Pea (*Pisum sativum* (L.)) var AP-3obtained from local market of Hoshiarpur were used in the present pot experimental study.

The soil samples were analysed for their physicochemical properties. The pot experiment was conducted by taking the unsterilized soils from fields of Hoshiarpur. Out of 15 isolated Strains of *Rhizobia* only 4 strains viz SS3, SS7, SS8 and SS12 were mixed with the seeds of Pea (*Pisum sativum* (L.)) var AP-3. Uninoculated seeds served as control. After 60 days of sowing, the plants were observed for number of nodules, fresh and dry weight of plants, length of plants, leghaemoglobin content (Wilson and Reisenauer, 1963), Nitrogenase activity (Hardy *et al.*, 1973) and percent Nitrogen content (Mckenzie and Wallace, 1954).

Results and Discussion

All the 4 isolates (SS3, SS7, SS8 and SS12) along with the uninoculated control were used to study their effects on various symbiotic parameters in Pea variety AP-3 (Fig 1).

All the *Rhizobium* cultures resulted in significant increase in nodulation as compared to uninoculated control. Among all these cultures, the plants inoculated with strains SS8 and SS12 were better than the other strains and showed maximum number of nodules (53 in plants inoculated with SS8 and 20 in plants inoculated with SS12).

Cultivar var AP-3 represented a good interaction with these *Rhizobial* isolates and enhanced the fresh and dry weight of plants as compared to uninoculated control too. These isolates of *Rhizobia* also exhibited maximum plant length over control (Table 1).

Table.1 Effect of inoculation of *Rhizobium* Isolates on Number of Nodules, Fresh Weight (g)/ Plant, Dry Weight (g)/ Plant and Length (cm)/Plant

***Rhizobium* Isolates**	**No. of nodules**	**Fresh Weight (g)/ Plant**		**Dry Weight (g)/Plant**		**Length of Plants (cm)**	
		Roots	**Shoots**	**Roots**	**Shoots**	**Roots**	**Shoots**
Control	4	0.67	1.33	0.07	0.85	20.3	17.0
SS-3	17	2.70	6.40	1.06	4.22	21.5	31.0
SS-7	10	2.41	5.40	1.02	4.18	17.4	34.0
SS-8	53	11.35	12.20	8.17	9.16	33.0	40.3
SS-12	20	4.49	6.67	2.23	5.18	27.0	36.2

Table.2 Effect of inoculation of Rhizobium Isolation on Leghaemoglobin content, Nitrogenase activity and Percent Nitrogen/Plant

Rhizobium Isolates	Leghaemoglobin content (mg/0.5 g nodules)	Nitrogenase activity(µM C_2H_2 reduced/plant)	Total Nitrogen content (%N)
Control	1.16	0.15	0.98
SS-3	3.76	0.40	1.82
SS-7	3.76	0.32	1.40
SS-8	8.96	0.65	3.22
SS-12	6.36	0.43	2.10

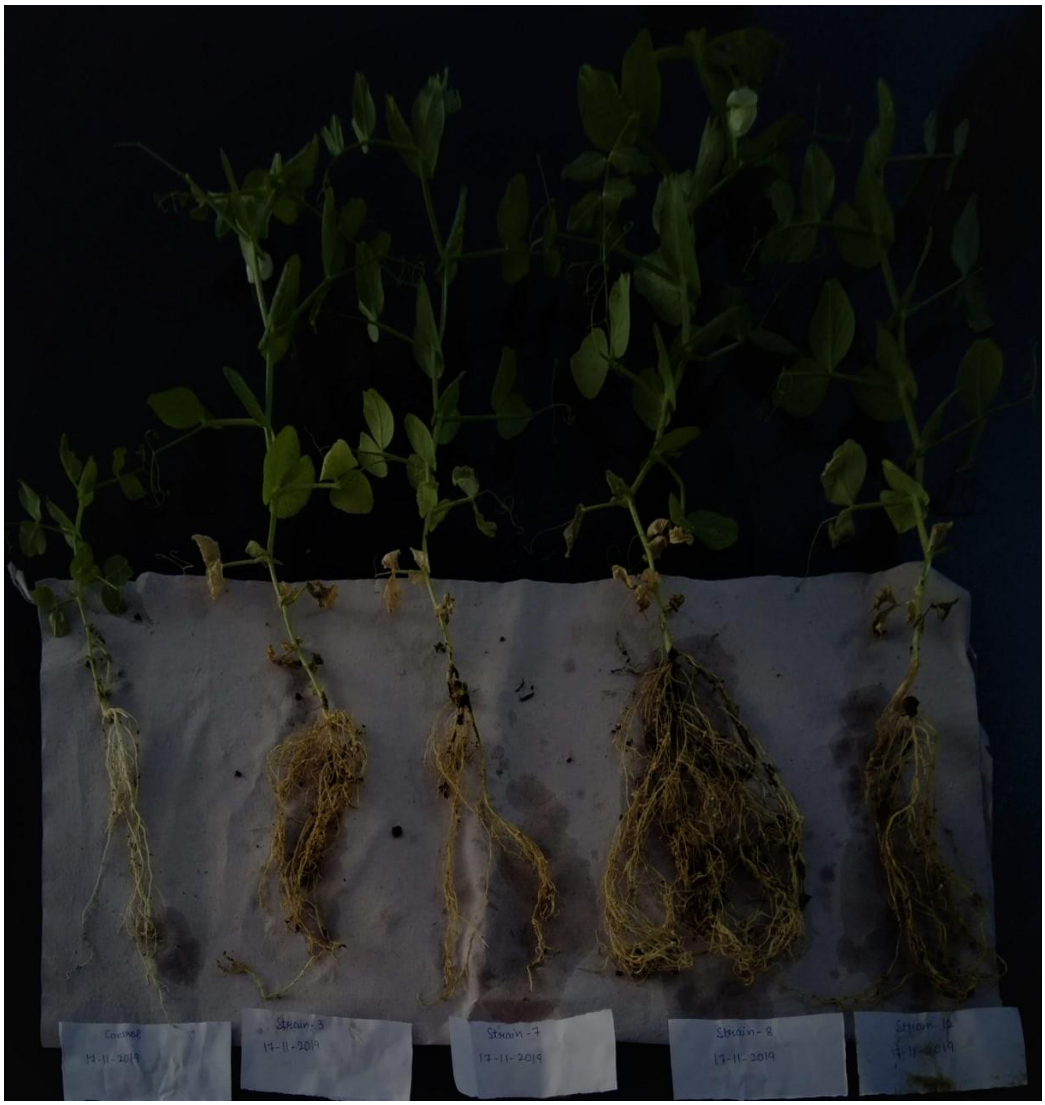

Figure.1

The Leghaemoglobin content of the nodules is taken as the index of nodule efficacy as it regulates the oxygen supply to the bacteroids and hence improves efficiency of nitrogenase. The Leghaemoglobin content in nodules inoculated by introduced *Rhizobium* isolates was found to be significantly high compared to uninoculated control. The nodules formed by inoculation of SS8 represented maximum leghaemoglobin content (8.96mg/0.5 g nodules) followed by SS12 (6.36mg/0.5 g nodules) and the least leghaemoglobin content was reported in plant nodules inoculated with SS7strain (3.76mg/0.5 g nodules).

Our results are in corrobation with the results of Sibbal *et al.,* (2001). Cultivar var AP-3 showed best interaction with all the isolates. The nitrogenase activity was reported to be highest in plants inoculated with strain SS8 (0.65µM C_2H_2 reduced/plant) followed by that of SS12 (0.43 µM C_2H_2 reduced/plant).Similar results were observed by Sibbal and Khurana (2002).

The nitrogen content of the plant is the whole sum of the nitrogen uptaken from the soil and that reduced by nodules from atmosphere. The maximum Nitrogen content of plant was reported by inoculation with SS8 (3.22%) and SS12 (2.10%) strains (Table 2). Our results are in consonance with the results of Chatli and Gupta (2014).

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