

Effect of *Rhizobium* Inoculation on Growth and Nodule Formation of Green Gram

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ABSTRACT

To find out the effect of *Rhizobium* inoculation and nitrogen on performance of mungbean (*Vigna radiata* L.), investigations were conducted at University of Arid Agriculture, Rawalpindi, during spring 2004. The research material consisted of mungbean variety (NM- 98) with treatments of seed and soil inoculation and nitrogen levels at 15, 30 and 45 kg ha⁻¹. Data were recorded on, number of nodules per plant, root length, plant height at maturity and biological yield per plant. Soil and seed inoculation in combination with N fertilizer positively affected the growth and nodule formation of green gram. Among all the treatments, seed inoculation + 15 kg N ha⁻¹ was found effective.

Key Words: *Rhizobium* sp; *Vigna radiata* L; Green gram; Growth; Nodule formation

INTRODUCTION

Greengram [*Vigna radiata* (L.) Wilezek] is a well-known pulse crop of Pakistan. It is a short duration crop and can be grown twice in a year. Being drought resistant, it can withstand adverse environmental conditions, and is successfully cultivated in rainfed areas. Mungbean is digestible, high in protein (22 - 24%; Malik, 1994) and does not cause flatulence that many other legumes do. Moreover, it is rich in vitamins as A, B, C, niacin, and minerals such as potassium, phosphorus and calcium, which are necessary for human body (Rattanawongsa, 1993). Owing to all these characteristics it is a good substitute of animal protein and forms a balanced diet when it is taken with cereals.

Rhizobium spp. invade the root hairs of mungbean and result in the formation of nodules, where free air nitrogen is fixed. These bacteria, although present in most of the soils vary in number, effectiveness in nodulation and N-fixation. It has been argued that usual native soil rhizobial populations are inadequate and are ineffective in biological nitrogen fixation. To ensure an optimum rhizobial population in the rhizosphere, seed inoculation of legumes with an efficient rhizobial strain is necessary. This helps improve nodulation, N₂ -fixation solicit crop growth and yield of leguminous crops (Henzell, 1988).

Although, this crop is capable of fixing atmospheric nitrogen through *Rhizobium* species living in root nodules, however, under our agro-ecological conditions, the nodulation of mungbean is poor and is a major cause of its lower yield. It was observed that inoculation of mungbean with *Rhizobium* spp. increased plant height, leaf area, photosynthetic rate and dry matter production (Thakur & Panwar, 1995). The present studies were conducted to evaluate effect of soil and seed inoculation alone and in combination with different nitrogen levels on nodule

formation and growth of mungbean plant and to compare the efficiency of seed inoculation against soil inoculation for nitrogen fixation.

MATERIALS AND METHODS

To evaluate the response of mungbean to *Rhizobium* inoculation and use of N-fertilizer, a pot experiment was conducted at University of Arid Agriculture; Rawalpindi. Mungbean variety NM- 98 was sown during spring on March 10, 2004 by using ten seeds per pot. The experiment was laid out in completely randomized design with three replications. Phosphorous was applied at 40 g pot⁻¹ in the form of single super phosphate. Three levels of nitrogen as 10, 20 and 30 g pot⁻¹ were applied in the form of urea. Both the fertilizers were applied at the time of sowing. For seed inoculation seeds were coated with paste of *Rhizobium* inoculum containing 10⁷ per g and sown in the pots. For soil inoculation 1 L solution of *Rhizobium* inoculum containing 10⁷ rhizobia per liter was added to the soil in each pot before sowing. Then seeds were sown in the inoculated pots. Five to six plants were maintained after germination in each pot and the treatments were replicated thrice. All agronomic practices were kept uniform and normal for all treatments. Twelve treatments as each seed and soil inoculation alone, nitrogen added to each of seed and soil inoculated plots at 15, 30 and 45 kg N ha⁻¹, and the same nitrogen rates were used alone. A control without inoculation and nitrogen application was included for comparison. Data on number of nodules per plant and root length at maturity, plant height at maturity, biological yield per plant (g) were recorded during the course of study by following standard procedures. The data were subjected to statistical analysis using Fisher's analysis of variance techniques and least significant difference test at 5% probability level was used

Table I. Effect of *Rhizobium* inoculation and nitrogen fertilizer on some traits of mungbean

Treatments	Number of nodules plant ⁻¹	Root length at maturity (cm)	Plant height at maturity (cm)	Biological yield (g plant ⁻¹)
Control	3.30 d	8.88 ab	13.70 d	3.21 f
Seed inoculation only	4.30 abc	9.77 a	13.91 d	3.55 f
Soil inoculation only	4.30 abc	7.93 bcd	16.16 cd	3.76 f
Seed inoculation+15 kg N ha ⁻¹	5.00 a	8.50 abc	20.76 ab	9.73 a
Seed inoculation+30 kg N ha ⁻¹	4.46 ab	6.96 cde	16.00 cd	6.33 bcd
Seed inoculation+45 kg N ha ⁻¹	4.00 bcd	5.70 e	18.20 bcd	7.33 bc
Soil inoculation+15 kg N ha ⁻¹	3.85 bcd	8.22 abc	17.53 bcd	6.26 bcde
Soil inoculation+30 kg N ha ⁻¹	3.90 bcd	6.80 cde	18.26 bcd	5.57 de
Soil inoculation+45 kg N ha ⁻¹	4.33 abc	6.10 e	22.56 a	4.53 ef
N at 15 kg ha ⁻¹	4.50 ab	6.42 de	20.30 abc	6.20 cde
N at 30 kg ha ⁻¹	3.51 cd	7.90 bcd	16.85 bcd	7.46 bc
N at 45 kg ha ⁻¹	3.80 bcd	6.05 e	16.06 cd	8.00 ab
LSD (0.05)	0.73	1.73	3.84	1.74

to compare the differences among treatment means (James *et al.*, 1997).

RESULTS AND DISCUSSION

Application of inoculation treatments and nitrogen fertilizer significantly affected number of nodules and the treatments significantly differed from each other as compared to the control (Table I). The highest number of nodules per plant (5.0) was observed in seed inoculation + 15 kg N ha⁻¹. Application of seed inoculation + 30 kg N ha⁻¹ and 15 kg N ha⁻¹ gave 4.46 and 4.50 nodules per plant, respectively and seed inoculation with nitrogen fertilizer applications showed better results than soil inoculation. Ramaswami and Oblisami (1986) reported the increase in nodules due to inoculation application. Increase in nodules per plant due to application of inoculation in combination with nitrogen fertilizer was also reported by Rashid *et al.* (1999).

Various treatments differed significantly with each other including control (Table I). The maximum root length (9.77 cm) was observed in response to seed inoculation alone and was significantly higher than seed inoculation + 15 kg N ha⁻¹, seed inoculation + 30 kg N ha⁻¹, soil inoculation + 30 kg N ha⁻¹, soil inoculation + 45 kg N ha⁻¹, 15 kg N ha⁻¹ and 45 kg N ha⁻¹ treatments, however, control, soil inoculation only seed inoculation + 15 kg N ha⁻¹, soil inoculation + 15 kg N ha⁻¹ and 30 kg N ha⁻¹ doses produced statistically same root length. The lowest root length (5.70) was recorded in soil inoculation + 45 kg N ha⁻¹ and this root length was significantly lower than control, seed inoculation only, seed inoculation + 15 kg N ha⁻¹ and soil inoculation + 15 kg N ha⁻¹ remained at par with it. These results are in line with the findings of Patra and Bhattacharyya (1998), who reported that seed inoculated plants exhibited significantly greater root and shoot length as compared to un-inoculated control plants.

Application of inoculation and nitrogen significantly affected plant height (Table I). Nitrogen fertilizer 45 kg ha⁻¹ the in combination with soil inoculation as well as 15 kg N ha⁻¹ alone and in combination with seed inoculation

increased the plant height over control, while other treatments were at par with it. Tallest plants (22.56 cm) were recorded in case of soil inoculation + 45 N kg ha⁻¹, however 15 kg N ha⁻¹ alone and in combination with seed inoculation remained at par with it. Seed and soil inoculation alone and in combination with various nitrogen doses did not show any significant difference for plant height as compared to control except seed inoculation + 15 kg N ha⁻¹ and soil inoculation + 45 kg N ha⁻¹ treatment. Hoque and Haq (1994) reported that inoculation of seed with *Rhizobium* significantly increase plant height of lentil. Harahap (1994) also reported that application of nitrogen fertilizer significantly affected plant height in mungbean. Khalil *et al.* (1989) observed that nitrogen alone or in combination with inoculation gave the maximum plant height in mungbean as compared to control.

Seed inoculation and nitrogen fertilizer significantly increased biological yield as compared to control (Table I) seed and soil inoculation alone and soil inoculation with 45 kg ha⁻¹ N failed to enhance biological yield over control. Seed inoculation + 15 kg N ha⁻¹ gave as good biological yield as was achieved with 45 kg N ha⁻¹ alone. All other treatments increased biological yield to variable extent over control. The increased biological yield in mungbean in all treatments might be attributed to high vegetative growth owing to the availability of nitrogen in soil and high moisture due to heavy rains during vegetative growth period.

On the basis of this study, it is concluded that inoculation of soil and seed with *Rhizobium* in combination with nitrogen fertilizer significantly affected the growth and nodules formation of green gram. Seed inoculation + 15 kg N ha⁻¹ performed better than other treatments and produced the maximum root nodules.

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