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Effect of Rhizobium Inoculation and Nitrogen Fertilization on Nodulation and Yield Response of Common Bean (Phaseolus vulgaries L.) at Boloso Sore, Southern Ethiopia

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Abstract

The cost of chemical fertilizer is high and their use efficiency especially nitrogen fertilizer is low. Use of biological nitrogen fixation as source of N and determining the optimum level of N as starter fertiliser are important option to increase productivity of legumes. A field experiment was conducted to assess the effect of rhizobium inoculation and N fertilizer application on nodulation and yield of common bean varieties in 2014 cropping season. Two common bean varieties (Omo-95 and Red Wolaita) were treated by three level of N (0, 23 and 46 kgNha^{-1}) and two rhizobium strains (BH129 and BH113). The treatments were arranged using randomized complete block design with three replications. The result of this study indicated that application of N fertilizer significantly suppressed the number of nodules while inoculation of rhizobium bacteria increased the number of nodules per plant significantly for the two varieties. Furthermore, both application of N fertilizer and inoculation of rhizobium increased hundred seed weight and seed yield ha^{-1}. Om-95 gave significantly higher number of seeds per pod and seed yield ha^{-1} than Red Wolaita. From this result, it can be conclude that inoculation of rhizobium enhanced nodulation of haricot bean varieties with better response of Omo-95 while application of N fertilizer had suppressing effect. Furthermore, both application of chemical fertilizer and inoculation of rhizobium increased yield of common bean with better performance of Omo-95 variety (2416.1 kgha^{-1}) treated by rhizobium strain called BH129. For better health of the soil and reducing cost of production, it is better to use seeds treated by bacteria but further study should be done by considering other soil and crop factors.

Keywords: Rhizobium, nitrogen, nodulation and yield

1. INTRODUCTION

Common bean(*Phaseolus vulgaries* L.) is a food legume that forms nodules with a diversity of rhizobial genotypes (Aguilar, *et al.* 2004): including *Rhizobium leguminosarum* bv. *Phaseoli*, *Rhizobium tropici*, *Rhizobium. etli*, *Rhizobium. gallicum* and *Rhizobium. giardinii* (Jordan, 1984; and Amarger *et al*., 1997). The percentage of biological nitrogen fixation of the N assimilation in common bean is lower as compared to other legumes being 40-50% compared to 75% with faba beans (*Vicia faba*), 70% with peas (*Pisum sativum*) and up to 95% with lupines (Werner, 1999). Hence, it is characterized with poor capacity to fix atmospheric nitrogen.

Rhizobia, large number of bacterial species, are able to fix atmospheric N due to a symbiotic relationship with legume plants. In a symbiotic relationship, both the plant and bacteria contribute to each other and benefit as a result of their association (Redmon and Smith, 2004). Different leguminous crops require specific rhizobium species for the formation of effective nodules and nitrogen fixation (Goss, *et al*., 2005). For new establishments, seed of legumes are required to be inoculated with the proper rhizobia specie. That is, the seed is coated with a sticking agent and the bacteria applied directly to the seed. At this places the bacteria is in close proximity to the seed at the time of germination. Infection of the plant by the bacteria takes place at the root hair, which indicates infection with a curling response. The plant's response to rhizobium infection is referred to as nodulation. Nodules appear as small lumps on the roots of legume plants. It is inside these nodules that N fixation occurs (Redmon and smith, 2004).

Nutrients play an essential role in increasing the seed yield of pulses. In legume, N is more useful because it is the main component of amino acid as well as protein (Hussain. *et al*, 2011). Bean growth and yield frequently benefit from the application of N fertilizer (Blaylock 1995). Added N often stimulates early bean growth because the first N that is fixed is used for nodule growth (Sprent and Thomas, 1984). Bean plants may be benefited from available soil N during pod filling if the ability of the plants to acquire N from symbiotic N_2 fixation is impaired by root disease or nodule senescence (Kumarasinghe *et al.,* 1992).

Although the use of mineral N fertilizer in production has increased substantially, its use(agronomic) efficiency is less than 50%. More than one-half of the applied N is lost through various processes (mainly run-off water and leaching), resulting in higher production costs and considerable environmental pollution (Ladha and Reddy, 1995). To solve the problem, two potential approaches have been explored. These are utilization of legumes that will efficiently fix atmospheric N and/or improvement on the uptake of native soil N and applied N by plants (Ladha *et al*., 1988a). Thus, this study was initiated to assess the effect of rhizobium inoculation and N fertilizer application on nodulation and yield of common bean varieties.

2. MATERIALS AND METHODS

2.1 Experimental Site: Field experiment was conducted on farmer's field during 2014 cropping season at Gurumo Koysha of Boloso Sore Woreda, Southern Ethiopia. The experimental site is located at an altitude of 2100 m.a.s.l, and having the average annual rainfall of 1750 mm. The major crops grown in the study area include maize (Zea mays), common bean(Phaseolus vulgaris), potato (Solanum tuberosum), sweet potato(Ipomea batatas), enset /false banana (Ensete ventricosum**)**

2.2 Treatments and Experimental Design: Treatments consisted of three levels of N fertilizer (0 23, and 46 kg ha^{-1}) and two rhizoium strains (HB 129 and HB 113). The common bean varieties (Red Wolia and Omo - 95) were used as test crops. The two bean varieties combined with ination. The treatments were laid out in a randomized complete block design (RCBD) with three three levels of N and two bacteria strains, as a result there were ten treatments comb replications. Each plot had a width and length of 3 m and spacing of 0.4, 1 and 1.5m were maintained between rows, plots and blocks, respectively. Seeds were inoculated with lignite-based inoculants of Rhizobium leguminosarum. All inoculants were done before planting under shade to maintain the viability of cells. Seeds were air dried for a few minutes before planting. The inoculated and non-inoculated seeds, as per treatment, were planted at the rate of 90 Kg ha^{-1} . As a precaution of cross-contamination, non-inoculated treatments were planted first followed by inoculated treatments. Phosphorus at rates of 46 kg P_20_5 ha^{-1} was applied to all plots uniformly at planting. Nitrogen was applied in the form of urea by spilt manner as half dose at planting and the remaining at vegetative stage. After germination, all crop management practices carried out during the growing period.

2.3 Soil Analysis: Before initiation of the trials representative soil samples were collected from depth of 0-30 cm at representative points of the experimental field. The samples were thoroughly mixed and composited to one representative sample for physio-chemical analysis. The sample was air dried and ground to pass through a 2 mm sieve to determine soil texture, pH, total nitrogen and available P. Soil pH was estimated by potentiometer method at soil: water ratio of 1:2.5. Total nitrogen by the micro kjeldhal method (Jackson, 1958) and available P was analyzed by Olsen method (Olsen *et al*., 1954). Soil particle size distribution was determined by hydrometer method (Day, 1965).

2.4 Data Collection and Analysis: Plant parameters recorded were plant height, pods per plant, seeds per pod, thousand seed weight (TSW), and grain yield. Plant height, pods per plant and seeds per pod were taken from five randomly selected plants per plot. Grain yield was harvested from central rows by avoiding border effects and converted to kgha^{-1} after adjusting the moisture content at 10%. Nodulation was assessed when 50% of plants of each plot exhibit flowering stage by carefully uprooting five plants randomly from each plot. The adhering soil was removed by washing gently over a metal sieve. The nodules from each plant were removed and separately spread on the sieve to drain water from their surface. Nodules were counted and their average was taken as number of nodules/plant

Collected data were subjected to analysis of variance using the general linear model SAS version 9.1 (SAS Inst., 2003). Treatments means were compared using the least significant difference (LSD) at 5% level of significance.

3. RESULT AND DISCUSSION

3.1 Physiochemical Properties of the Soil: The result of laboratory analysis of soil sample taken before sowing indicated that the chemical environment of the site was strongly acidic (pH=5.4). The available phosphorus content of the soil was found at low range(8.4 ppm) and also it had total nitrogen of 0.16% which is found at medium rang as per the rate of Landon (1991). Textural class of the soil was sandy clay loam.

3.2 Nodulation: Application of nitrogen fertilizer had significant ($P<0.05$) effect on number of nodules (Fig 1). As the application of N increased the number of nodules decreased and as compared with plants treated by N fertilizer, the control had higher number of nodules. This result indicated that application of N had suppressing effect on nodulation which might be attributed to the inhibitory effect of nitrate (NO_3-) since higher concentration of nitrate depresses nodulation of legumes (Zahran, 1999; Cheema and Abrar, 2000). In line with the result of this study Dogra and Dudeja (1993) reported that application of N had depressed the number of nodules of different grain legumes. So application of nitrogen for increasing legumes productivity may be encouraged for soil with low fertility especially those with low N concentration (Zahran, 1999).

Number of nodule per plant was significantly ($P<0.05$) influenced by interaction effect of varieties with rhizhobium inoculation and N application (Fig. 1). The highest and the lowest number of nodules per plant were recorded from Omo-95(V1) treated by rhizobium strain called *HB 129* and Red Wolaita(V2) which were treated by 46 kgNha^{-1}, respectively. Inoculation of rahizobium strains increased the number of nodules significantly for both varieties. From the two raizhbium strain, BH129 resulted in significantly higher number of nodules for Omo-95 than Red Woliata while BH113 resulted equal number of nodule per plant for both varieties (Fig. 1).

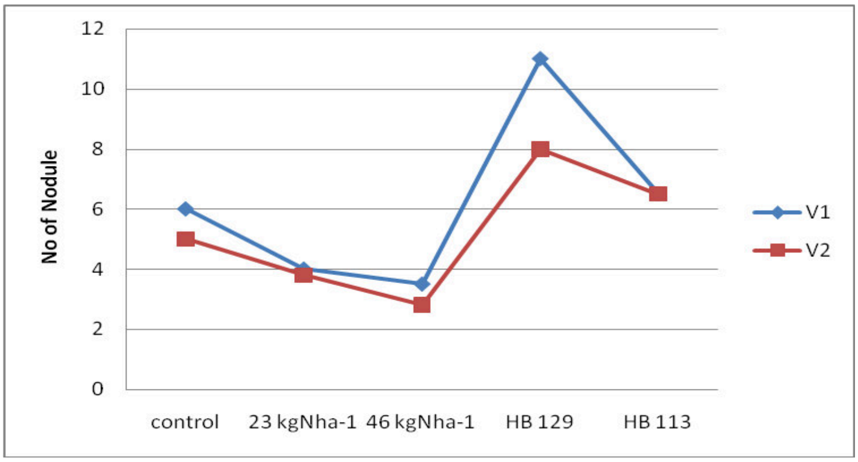

Figure 1. Effect of N application and Rahizobium inoculation on nodule number of common bean varieties.

Increasing of nodule number when the seeds treated by rahizobium strain indicated that the native rahizobium population density nodulating the legume might not be adequate and this in turn brought response of the plant to inoculation of the rahizobium strains (chemining'wa *et al.*,2007). Similarly, Otieno *et al.* (2007) reported that rhizobium inoculation significantly increase the number of nodules per plant.

3.3 Yield and Yield Components: Inoculation of rhizhobium strain and application of N didn't significantly ($P<0.05$) influenced pod and seed number while hundred seed weight and seed yield ha^{-1} were significantly influenced (Table 1). The maximum and minimum hundred seed weight and seed yield ha^{-1} were recorded from plants treated by inoculation of rhazobium strain called BH 129 and control, respectively. Interaction effects of varieties with rhizobium inoculation and N application was nonsignificant ($P<0.05$).

This study indicated that application of N fertilizer enhance yielding of haricot bean varieties since the concentration of total nitrogen of soil(0.16%) in the experimental site wasn't at optimum range for plant growth. This condition may resulted the plant to respond for the applied N that the plant use especially for early growth initiation and for better pod formation (Sprent and Thomas 1984, Kumarasinghe *et al.,* 1992 and Blaylock 1995). In line with this result, Otieno *et al.* (2007) reported that application of fertilizers increase yield and yield component of crop legumes.

Table 1. Effects of Rhizobium inoculation and N application on yield and yield component of common bean varieties

	N rates	Pods/plant	Seeds/pod	HSW	Seed yield (kgha^{-1}
Variety mean	Red Wolaita	10.4	3.9333	3.6	1845.2
	Omo-95	13.3	5.333	4.3	2284.3
	LSD	NS	0.5384	NS	375.4
N rates mean	0	9.767	4	3.29	1468.1
	23	11.6	4	3.51	2160
	46	13.867	4.7	3.47	2103.3
	HB 129	13.767	4.2	5.62	2416.1
	HB113	10.5	3.8	5.11	1876.3
	LSD	NS	NS	1.62	593.57
	CV (%)	18.98	6.98	23.25	23.37

HSW=Hundred seed weight, HB129 and HB113 are rhizobium strains called *HB 129 and HB 113*, respectively.

The significant yield response of plants to inoculation of rhizobium may be attributed to better nodule formation than the control which enhance better fixation of atmospheric nitrogen and in turn yield formation. Otieno *et al.* (2007) reported that inoculation of bacteria increases yield response of some food legumes. There was no significant ($P<0.05$) difference between the two varieties on yield component parameters but seed per pod and seed yield ha^{-1} were significantly varied (Table 1). Omo-95 gave the higher seed yield than Red Wolaita which may be attributed to the variability on yielding potential of the two varieties.

4.CONCLUSION AND RECOMMENDATION

From this result, it can be concluded that inoculation of rhizobium increased nodulation of common bean varieties with better response of Omo-95. Furthermore, both application of chemical fertilizer and inoculation of rhizobium increased seed yield ha^{-1} with better performance of Omo-95 treated by rhizobium strain called BH129. For better health of the soil and reducing cost of production, it is better to use seeds treated by bacteria but further study should be done by considering other soil and crop factors.

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