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Effect of Cowdung, NPK and *Rhizobium* Inocula on Growth and Soil Status of Summer Mungbean (*Vigna radiata*) in Acid Soil

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Authors' contributions

This work was carried out in collaboration between all authors. Author KY conducted the study, performed statistical analysis, searched literature and wrote drafts of the manuscript. Author MAA supervised, guided and managed overall study. Author MAK designed the experiment, guided, supervised and forwarded all drafts of manuscript which was written by author KY. All authors read and approved the final manuscript.

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ABSTRACT

This research was carried out to evaluate the effect of cowdung, NPK and *Rhizobium* inocula on growth and soil status of summer mungbean (*Vigna radiata*) on acid soil of Khadim Nagar, Sylhet, Bangladesh. Seven treatments; Control (without *Rhizobium* inocula and fertilizers), Cowdung (CD), CD+RI, NPK, NPK+RI, ½(NPK)+RI and *Rhizobium* inocula (RI) were built into a randomized complete block design with three replications. The rate of Urea-N, TSP-P and MoP-K were 14, 14 and 17.5 kg ha^{-1}, respectively. Cowdung was 8 tons hectare and RI used @ 45 g kg^{-1} seed. The randomly selected 5 plants were uprooted at 30, 45, 60 and 90 days after sowing for growth characters analyses. The initial and post-harvest soils were collected and analyzed for pH, organic matter, total N, exchangeable K, available P, available S and available B. Cowdung, NPK and *Rhizobium* inocula had significant effects on growth characters of Mungbean. Sole application of cowdung @ 8 tons hectare produced longer root with taller plants, higher number of branches

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plant^{-1} along with longer and wider trifoliate leaves which highly influenced the biomass of Mungbean. NPK fertilizers and *Rhizobium* application alone or in combined form did not significantly affect the plant growth over sole application of cowdung. The heavy rainfall also limits the NPK and *Rhizobium* activity that affects the plant growth. The post-harvest soils status was not significantly influenced due to treatments application. It may be concluded that cowdung nutrients supply were more efficient than that of NPK and *Rhizobium* fertilizers in acid soils.

Keywords: Vigna radiata; soil pH; cowdung; inocula; N_2 fixation.

1. INTRODUCTION

The soils of Sylhet region in Bangladesh are acidic and stand as one of the major constraints of crop production in the area; as major agricultural crop in Bangladesh do well in neutral pH soils. Studies have shown that Mungbean plant grow well in a neutral, well aerated soils amended with manure rich in carbon contents for symbiotic relationship between root nodules and soil organisms. Soil acidity constrains symbiotic N_2 fixation in both tropical and temperate soils, limiting *Rhizobium* survival and persistence in soil and reducing nodulation [1,2]. The nitrogenase enzyme and nif gene activity of *Rhizobium* was reduced because the bacteria are aerobic and prefer neutral pH. An antagonistic behavior of soil microorganisms was observed for N_2 fixation in acid based problem soils.

Mungbean is usually cultivated in two seasons of Bangladesh i.e. Rabi and Kharif-1. Rabi season spread from October-November and Kharif-1 season begins in February and ends in March. Recently a number of summer varieties are introduced by Bangladesh Agricultural Research Institute (BARI) and Bangladesh Institute of Nuclear Agriculture (BINA). This summer varieties are very popular in the Southern, Western and Northern part of Bangladesh; but is not popular in Sylhet region due to high rainfall and acidic nature of the soil. In Bangladesh, the total production of winter Mungbean was 0.116 million M tons with an average of 960 kg ha^{-1} under 0.121 million ha land. Whereas, the total production of summer Mungbean was 0.065 million M tons with an average of 1250 kg ha^{-1} under 0.052 million ha land [3]. Mungbean cultivation in acid soil may be adopted by applying manures and raising soil pH for improving such problem soils. The aim of this research was to evaluate the effect of cowdung, NPK and *Rhizobium* inocula on growth and soil status of summer Mungbean in acid soil.

2. MATERIALS AND METHODS

The experiment was conducted at the farm of Agricultural Training Institute (ATI), Khadim Nagar, Sylhet in Bangladesh during kharif-1 on 26 April to 31 July in 2014. The soil belongs to the Brown Hill soils under agro-ecological zone of Northern and Eastern Piedmont Plains (AEZ-22). The soil was acidic with loamy sand textural class and pH of 5.21. The test variety was Binamoog-8 of *Vigna radiata.* The treatments were Control (without *Rhizobium* inocula and fertilizers), Cowdung (CD), CD+RI, NPK, NPK+RI, ½(NPK)+RI and *Rhizobium* inocula (RI). The experiment was laid out in a randomized complete block design (RCBD) with three replications. Cowdung was applied @ 8 t ha^{-1}, the nutrients of N, P and K were applied at 14, 14 and 17.5 kg ha^{-1} from the sources of urea, TSP and MOP, respectively. The nutrient composition of cowdung was N (0.85%), P (0.32%), K (0.26%), S (0.21%) and organic carbon was 8.1%. The seed inoculation was done by coating the seeds with mixtures of the molasses (20 g kg^{-1}) and *Rhizobium* inocula (45 g kg^{-1} seed). The net plot size was 3 m × 2 m with 1 m proper drainage channel made surrounding the plots. The seed rate was 30 kg ha^{-1} at 25 cm × 10 cm plant spacing. The un-inoculated seeds were sown after the inoculated seeds on the same day. The intercultural operations and plant protection measures were taken when needed; especially the drainages done 5 times and the roofing was done with polythene sheet at early seedling stage of Mungbean to protect it from excessive rain water. Data on growth were recorded by uprooting five plants randomly from each plot at 30, 45, 60 and 90 DAS (final harvest). Plant roots were separated from shoot parts to measure root and shoot length in centimeter from each of the individual 5 plants. The total number of trifoliate leaves and branches were counted from 5 selected representative plants and average was obtained into per plant. The 10 trifoliate leaves of different sizes identified from each of 5 selected plants to measure leaf length and breadth in centimeters. The leaf area was calculated by measuring average length and breadth of 10 trifoliate leaves from individual 5 plants of per plot. Leaf area (cm^2 per plant) = K × average length × average breadth. Where, K = Kemp's constant (for dicot leaves = 0.66) [4]. Immediately after shoot collection of 5 selected plants, the fresh weight (g) was taken by electric balance and converted into tons per hectare. Then the samples were dried by electric oven at 70℃ for 48 hours, weighed and converted of tons per hectare. The initial and post-harvest soils samples were collected and processed. The soils were analyzed for soil pH by Glass electrode pH meter method [5], organic matter was determined by Wet oxidation method [6], total Nitrogen was measured by Micro-Kjeldahl method [7], exchangeable Potassium was measured by Flame Photometer [8], available Phosphorus was determined by Bray-1 method [9], Sulfur [10] and Boron [11] were identified by Spectrophotometer method. The mean was calculated and analysis of variance was performed by F-test and mean differences were adjudged by Duncan's Multiple Range Test [12].

3. RESULTS AND DISCUSSION

3.1 Growth Characters

3.1.1 Root length

Root length of Mungbean was significant (P <0.01) due to treatments (Table 1). The highest root length was recorded from the treatment of cowdung at each stage which ranged from 9.20 cm at 30 DAS to 12.36 cm at 90 DAS. Whereas, the lowest root length of 6.74 cm at 30 DAS to 7.92 cm at 90 DAS was recorded due to control except at 60 DAS. Nutrients efficiency of cowdung was higher than chemical fertilizers which allowed the production of deep rooted and also enhanced root proliferation. The findings are agreed with that of Hannan et al. [13], Otieno et al. [14], Karthikeyan et al. [15] and Abe et al. [16] who stated that manure allows better root growth and hence promotes better nutrient uptake.

3.1.2 Plant height

A significant (P <0.01) response was observed in plant height of Mungbean due to different treatments (Table 1). The height of Mungbean ranged from 13.22 at 30 DAS cm to 62.77 cm at 90 DAS. The tallest plant recorded mean values of 24.61 cm at 30 DAS and 62.77 cm at 90 DAS, produced at each stage due to cowdung treatment application. The shortest plant was produced due to control at each stage which ranged from 13.22 cm at 30 DAS to 21.71 cm at 90 DAS. Cowdung provided all essential nutrients to soil for plant uptake which supported the findings of Polthanee et al. [17] who reported that cattle manure applied at the highest rate of 9375 kg ha^{-1} had the highest plant height. A negative effect of *Rhizobium* was observed due to acid [18] with coarse textured soil [19] and moist condition.

3.1.3 Leaf number

The leaves per plant vary significantly (P <0.01) due to application of treatments (Table 2). The leaves per plant were 5.87 at 30 DAS and 7.94 at 90 DAS, recorded at each stage in cowdung applied plots. Whereas, leaves per plants of

Table 1. Effect of cowdung, NPK and *Rhizobium* inocula on root length (cm) and plant height (cm) Plant^{-1} at 30, 45, 60 and 90 DAS

Treatment	Root length (cm plant^{-1})				Plant height (cm plant^{-1})			
	30 DAS	45 DAS	60 DAS	90 DAS	30 DAS	45 DAS	60 DAS	90 DAS
Control	6.74b	6.82c	7.55d	7.92b	13.22c	15.66d	19.02d	21.71d
CD	9.20a	11.25a	12.04a	12.36a	24.61a	54.27a	61.49a	62.77a
CD+RI	7.47b	11.14a	11.28a	11.33a	22.83a	48.24ab	58.91a	58.58a
NPK	7.61b	10.05ab	9.26b	9.40b	18.30b	39.64b	45.61b	48.19b
NPK+RI	7.06b	8.44bc	8.96bc	9.07b	17.42bc	29.18c	38.63b	44.03b
½(NPK)+RI	7.05b	7.35c	7.80cd	8.16b	18.51b	24.51cd	29.57c	33.82c
RI	6.87b	7.03c	7.43d	7.98b	14.60bc	17.31d	19.52d	23.19d
Level of significance	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
CV (%)	7.85	10.28	5.65	7.28	8.78	11.95	8.18	6.64
SE	0.337	0.527	0.299	0.398	0.938	2.256	1.839	1.602

In a column figure(s) bearing common letter(s) do not differ significantly whereas figure(s) bearing dissimilar letter(s) indicate significantly different by DMRT. Here, CD = Cowdung, RI = Rhizobium inocula, SE = Standard error, DAS = Days after sowing

3.47 at 30 DAS and 3.50 at 90 DAS were produced in control plots. Affluent plant growth due to cowdung was produced higher number of branches with trifoliate leaves plant^{-1} which also agreed to the findings of Umunna and Anselem [20].

3.1.4 Branch number

Application of different fertilizers significantly (P <0.01 and P <0.05) affected branches number plant^{-1} of Mungbean (Table 2). The highest number of branches were recorded from cowdung applied treatment at each stage which ranged from 2.60 at 30 DAS to 4.80 plant^{-1} at 90 DAS. Whereas, the lowest number of branches plant^{-1} were obtained from the control plot except at 90 DAS. The similar result was attributed to the findings of Umunna and Anselem [20] who stated that kitchen ash produced higher branches and leaves plant^{-1} of Mungbean.

3.1.5 Leaf area

Leaf area of mungbean was significantly (P <0.01) affected due of application of different fertilizers (Table 3). The highest leaf area was observed in cowdung treated plots. On the contrary, the lowest leaf area was obtained in control plots.

Table 2. Effect of cowdung, NPK and *Rhizobium* inocula on the leaves and branches number plant^{-1} at 30, 45, 60 and 90 DAS

Treatment	Leaves number (Number plant^{-1})				Branches number (Number plant^{-1})			
	30 DAS	45 DAS	60 DAS	90 DAS	30 DAS	45 DAS	60 DAS	90 DAS
Control	3.47c	4.14c	4.64d	3.50c	1.50c	1.60c	1.94c	2.17c
CD	5.87a	8.92a	9.74a	7.94a	2.60a	2.74a	4.17a	4.80a
CD+RI	5.07ab	8.87a	9.60a	6.94ab	2.07b	2.34ab	3.17b	3.67ab
NPK	4.10ab	7.27ab	7.57b	5.24bc	2.14ab	2.20ab	3.07b	3.47b
NPK+RI	4.60b	6.80b	7.10bc	4.44c	2.07b	2.07bc	2.47bc	2.74bc
½(NPK)+RI	4.34bc	5.74bc	6.14bcd	3.57c	2.07ab	2.07bc	2.14c	2.20c
RI	4.00bc	5.07c	5.44cd	3.77c	1.84bc	1.94bc	2.00c	2.07c
Level of significance	0.01	0.01	0.01	0.01	0.05	0.05	0.01	0.01
CV (%)	9.09	9.91	9.79	16.46	13.55	14.12	12.40	15.48
SE	0.242	0.383	0.406	0.479	0.159	0.174	0.194	0.269

In a column figure(s) bearing common letter(s) do not differ significantly whereas figure(s) bearing dissimilar letter(s) indicate significantly different by DMRT. Here, CD = Cowdung, RI = Rhizobium inocula, SE = Standard error. DAS = Days after sowing

Table 3. Effect of cowdung, NPK and *Rhizobium* inocula on leaf area (cm^2 per plant) of mungbean at 30, 45, 60 and 90 DAS

Treatment	Leaf area (cm^2 per plant)			
	30 DAS	45 DAS	60 DAS	90 DAS
Control	13.27d	20.80e	28.32e	31.53c
CD	55.87a	75.33a	85.15a	89.19a
CD+RI	46.31b	64.52b	72.05ab	77.78a
NPK	38.96bc	52.99c	57.65bc	58.65b
NPK+RI	35.63c	51.50c	56.99bc	59.12b
½(NPK)+RI	32.26c	41.00d	46.83cd	48.91b
RI	13.50d	27.21e	31.90de	33.04c
Level of significance	0.01	0.01	0.01	0.01
CV (%)	11.44	7.87	11.33	9.05
SE	2.197	2.162	3.540	2.974

In a column figure(s) bearing common letter(s) do not differ significantly whereas figure(s) bearing dissimilar letter(s) indicate significantly different by DMRT. Here, CD = Cow dung, RI = Rhizobium inocula, SE = Standard error. DAS = Days after sowing

3.1.6 Fresh weight

Application of different fertilizers had significantly (P <0.01) effect on fresh weight tons per hectare of Mungbean plant (Table 4). The highest fresh weight tons per hectare was recorded in cowdung treated plants and the lowest fresh weight was recorded in the control plots. Higher availability of nutrients in organic fertilizer was the main factor contributing to higher biomass of plants stated by Singh and Agrawal [21].

3.1.7 Dry weight

A significant (P <0.01) response was observed on oven dry weight tons per hectare due to application of different treatments (Table 4). The highest and the lowest oven dry weight plant^{-1} were observed in cowdung and control, respectively. Cowdung influenced production of higher Mungbean biomass than other treatments including control.

3.2 Effect of Cowdung, NPK and *Rhizobium* Inocula on Soil Chemical Properties

The post-harvest soil nutrients were not significantly improved due to different fertilizers application (Tables 5 and 6). Cowdung and CD+RI application slightly increased the values of soil pH, soil organic matter, total nitrogen, exchangeable potassium, available phosphorus and available S higher than other treatments

Table 4. Effect of cowdung, NPK and *Rhizobium* inocula on fresh weight and oven dry weight tons per hectare at 30, 45, 60 and 90 DAS

Treatment	Fresh weight (tons/hectare)				Oven dry weight (tons/hectare)			
	30 DAS	45 DAS	60 DAS	90 DAS	30 DAS	45 DAS	60 DAS	90 DAS
Control	0.829d	1.118d	1.351f	1.407e	0.136d	0.303d	0.561d	1.031c
CD	2.194a	3.190a	4.901a	5.987a	0.700a	1.847a	2.648a	3.129a
CD+RI	1.619b	2.964a	4.458b	4.779b	0.317b	1.227b	1.922b	2.836a
NPK	1.574b	2.532ab	3.486c	3.598c	0.237bc	0.874c	1.178c	1.804b
NPK+RI	1.396bc	2.034bc	2.950d	3.413c	0.202cd	0.826c	1.070c	1.261bc
½(NPK)+RI	1.058cd	1.661cd	2.076e	2.274d	0.209cd	0.453d	0.721d	1.167c
RI	0.839d	1.173d	1.386f	1.407e	0.168cd	0.334d	0.617d	1.142c
Level of significance	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
CV (%)	13.24	12.59	4.82	9.93	11.50	12.87	10.02	13.68
SE	0.298	0.438	0.239	0.538	0.0548	0.178	0.207	0.401

In a column figure(s) bearing common letter(s) do not differ significantly whereas figure(s) bearing dissimilar letter(s) indicate significantly different by DMRT. Here, CD = Cowdung, RI = Rhizobium inocula, SE = Standard error. DAS = Days after sowing

Table 5. Effect of cowdung, NPK and *Rhizobium* inocula on pH, organic matter, total nitrogen and Exchangeable K of post-harvest soil

Treatment (Post-harvest)	Soil pH	Soil organic matter (%)	Total N (%)	Exchangeable K (meq100g $^{-1}$)
Control	5.205	1.260	0.138	0.122
CD	5.242	1.273	0.149	0.123
CD+RI	5.240	1.273	0.149	0.124
NPK	5.224	1.269	0.147	0.123
NPK+RI	5.223	1.267	0.147	0.123
½(NPK)+RI	5.217	1.264	0.145	0.122
RI	5.210	1.261	0.141	0.122
Level of significance	NS	NS	NS	NS
CV (%)	1.06	3.56	14.25	14.09
SE	0.032	0.026	0.012	0.009
Initial soil	5.21	1.26	0.141	0.123

In a column figure(s) do not bearing any letter(s) indicate the treatments are insignificant. Here, CD = Cowdung, RI = Rhizobium inocula, SE = Standard error. NS = non significant

Table 6. Effect of cowdung, NPK and *Rhizobium* inocula on available P, S and B of post-harvest soil

Treatment (Post-harvest)	Available P	Available S	Available B
		μg g^{-1} of soil	
Control	11.177	9.433	0.113
CD	11.426	9.477	0.117
CD+RI	11.447	9.473	0.117
NPK	11.412	9.443	0.116
NPK+RI	11.381	9.450	0.116
½(NPK)+RI	11.380	9.440	0.116
RI	11.191	9.440	0.116
Level of significance	NS	NS	NS
CV (%)	4.37	5.44	34.63
SE	0.287	0.297	0.023
Initial soil	11.18	9.45	0.118

In a column figure(s) do not bearing any letter(s) indicate the treatments are insignificant. Here, CD = Cow dung, RI = Rhizobium inocula, SE = Standard error. NS = non significant

including the control. The slight increase in values of the soil properties mentioned could be attributed to the fact that soil nutrients were slowly mineralized in cowdung fertilizer and its residual effects were not affected by the action of rain water (leaching), hence improved soil health. This agreed to the submission of Bhriguvanshi [22] who observed that some of plant nutrients, when added to the soil in the inorganic form have low efficiency as compared with the effect of same nutrients applied along with organic manures and Bocchi and Tango [23] stated that positive response of manure on soil and legume due to the quantity of manure N already available for the plants, manure releases N, P, K and other micronutrients through mineralization process. The excessive rainfall recorded in the study area which occurred between May and July of the study year might have increased the leaching processes of the nutrients released into the soil by the amendments, hence, reduced values of the post-harvest soil nutrients.

4. CONCLUSION

In acid soil Mungbean showed better responses to cowdung fertilizer rather than NPK fertilizer and *Rhizobium* biofertilizer. The post-harvest soil values were slightly increased due to CD and CD+RI. The performance of chemical fertilizers were retarded and this may be due to high rainfall and soil pH. The poor function of *Rhizobium* inocula was observed because soil and environmental limitations stopped N_2 fixation by microbes which later competed with crops for food sources in the soil.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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