EFFECT OF VARIETIES AND BIOFERTILIZERS ON GROWTH AND YIELD OF MUNG BEAN IN KAILALI, NEPAL

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ABSTRACT

Mung bean, one of the most important crop in terms of food and nutritional security, is often neglected in terms of soil nutrient management in cereal based cropping system, where biological properties of soil is ignored and application of chemical fertilizers is at large. Hence this study was conducted at Agronomy Farm, Far Western University, Kailali, Nepal during April-June 2022 to evaluate varieties under different biofertilizers and their combination on vield and vield attributing traits of mung bean. The experiment was laid out in factorial randomized complete block design (RCBD) with three replications. The treatment consisted of 3 varieties of mung bean viz. Pratigya, Kalyan and SML-668 and 6 levels of biofertilizers viz. Rhizobium (R), Rhizobium+ Trichoderma (RT), Rhizobium+Pseudomonas (RP), Rhizobium+Trichoderma+Pseudomonas (RTP), Trichoderma+Pseudomonas (TP) and control. Growth and yield of mung bean were significantly influenced by variety, biofertilizers and their combinations but no significant interaction effects were observed. SML-668 performed better over Pratigya and Kalyan respectively in root parameters, leaf area index, yield attributing traits and yield generating 1.81 t/ha grain yield. Similarly, the combination of biofertilizers (RTP) generated significantly highest grain yield followed by RP and RT. The grain yield obtained with application of Rhizobium (R) was statistically at par with that from TP. The control plot yielded significantly lesser grain yield which was 7.6% lesser than rhizobium (R). So, the use of SML-668 variety and application of rhizobium either alone or in combination is recommended for better yield of mung bean at similar condition under rice-wheat cropping system.

Keywords: Rhizobium, Pseudomonas, Trichoderma, effective nodules, grain space

INTRODUCTION

Mung bean (Vigna radiata), commonly known as green gram, is important grain legumes grown across South Asia. The grain of mungbean contains 25-26% protein, 3% vitamin,51% carbohydrates and 4% mineral (Afzal et al., 2008). This crop is cultivated on approximately 7.3 million hectares' land across the globe, with India and Myanmar each accounting 30% of total production of 5.3 million metric tons (Nair &Schreinemachers, 2020). More than 75% of mung bean cultivated in Nepal is produced from the Eastern and Central region, whereas only 25% is produced in the Western terai and foothills (Neupane et al., 2006). This pulse crop is mostly grown during hot summer months in Nepal for its high quality protein in grains and its nitrogen fixing ability in soils. Besides, this crop act as a break to insect-pest and disease cycle in cereal based cropping system (Kumar et al., 2018). Due to its short life cycle this crop could be grown in the fields with slight irrigation immediately after wheat crop, which otherwise remains fallow, during the summer months. Despite having important role in improving soil fertility and productivity of the rice-wheat cropping system, the area under this crop is declining.

Green gram cultivated in 8266 ha land produced 10468 tons with productivity of 1.26 t/ha has been reported in a study (Darai et al., 2021). Though the average yield of legumes has increased from 0.95 t/ha to 1.26 t/ha in the past 10 years, the area under legumes has declined (MoALD, 2023). Unavailability of quality seeds, lack of mechanization for harvesting and threshing, free grazing practices and improper nutrient management especially N has been identified as major constraints to mung bean cultivation in the far western terai region of Nepal. Moreover, intensive rice-wheat cropping system has promoted excessive use of chemical fertilizers, which in turn has deteriorated soil physical, chemical and biological properties making it difficult to get better yield from the leguminous crops grown in between. So loosening the soil to greater depth, balanced application of chemical fertilizers, organic fertilizers as well as addition of biofertilizers is equally important for improving soil fertility as well as improving the growth and yield of mung bean.

Biofertilizers are cost effective and eco-friendly (Barman et al., 2017) biological preparations of effective microorganisms (Singh et al., 2011), which replenishes and restores long term soil fertility through their biological activities in the rhizosphere thus encouraging antagonism and control phytopathogenic organisms (Silpa et al., 2021). Several biofertilizers has been reported to improve soil fertility by solubilizing insoluble

phosphates, improving nitrogen fixation through enhanced nodulation and producing plant growth promoting substances (Borkar, 2015; Htwe et al., 2019), thus increasing the yield of field crops by 10-40% (Daniel et al., 2022). Rhizobium, a gram negative rod shaped bacteria (Mishra et al., 2013)has been reported to increase the yield of grain legumes significantly by improving the root biomass (Rehman et al., 2019)through formation of nodules (Bhat et al., 2015)and converting nitrogen to ammonia throughout the growth stages (Sneha et al., 2018). Rhizobium bacteria fixes 33-643 Kg/ha nitrogen in soybean,125–143 Kg/ha nitrogen in black gram whereas 25–100 kg Kg/ha nitrogen in green gram (Macik et al., 2020; Gopalakrishnan et al., 2015). Application of Pseudomonas bacteria, has been reported to induce systemic drought tolerance in moong bean (Kumari et al., 2016). Others have reported increase in shoot growth (Egamberdieva et al., 2013) and yield of legumes through IAA production and root colonization Malik and Sindhu (2008) due to application of Pseudomonas. Similarly, Trichoderma, also considered a biofungicide (Verma et al., 2007) has reportedly improved the growth of plants by facilitating the production of growth-promoting compounds like IAA, cytokinins, gibberellins, zeatin and auxin (Contreras-Cornejo et al., 2016)., improving nutrient solubilization in rhizosphere, secreting antifungal substances to fight against phyto pathogens (Kumar et al., (2020) and inducing tolerance to water stress (Mastouri et al., 2012).

Inspite of having its significant role, its use by Nepalese farmers in leguminous crops including mung bean, is limited. Few researches has been done to study the effectiveness of different biofertilizers on recommended mung bean varieties in Nepal. Thus, this research seeks to address this research gap by evaluating the comparative advantages of employing biofertilizers on mung bean. The findings of this study will be invaluable to farmers interested in expanding their mung bean cultivation on a larger scale.

MATERIALS & METHODS

Experimental site

The field experiment was carried out during April-June, 2022 at Agronomy farm, Far Western University, Tikapur-1, Kailali to study the effects of different biofertilizers on the growth and yield of mung bean varieties. The experimental site situated 28°32' 26" N latitude, 81°07' 24" E longitudes at an elevation of 159 masl had sandy loam soil. The soil had 1.45 % organic matter content, 0.07 % nitrogen, 17.49 kg/ha phosphorous and 108 kg/

ha potassium and pH 6.6. The average maximum and minimum temperatures during the experiment period was 38.7°C and 23.11°C respectively. The lowest temperature during the growth period was during germination stage (16.5°C) and the highest (40.51°C) was during the maturity stage. Similarly, the site received a total of 46.05 mm precipitation with the highest of 23.1 mm at maturity stage as shown in Figure 1.

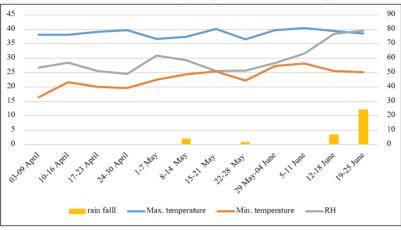
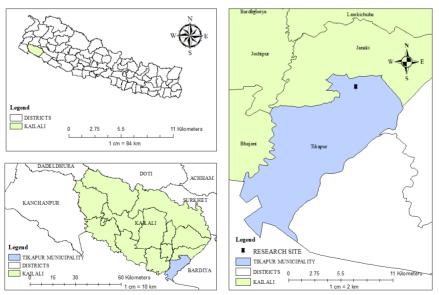


Figure 1. Weather condition during the experiment at Tikapur-01, Kailali, Nepal, 2022 *(Source: Department of hydrology and meteorology, Babarmahal, Kathmandu, Nepal)*



STUDY AREA MAP SHOWING RESEARCH SITE, TIKAPUR MUNCIPALTY, KAILALI, NEPAL

Figure 2. GIS map of study area showing research site, Tikapur muncipalty-01, Kailali, Nepal

Source: Extracted from New political and administrative boundaries Shape file of Nepal (GIS ARC MAP 10.4)

Experimental details

The experiment was laid out in Factorial RCBD (randomized complete block design) with three replications. The factors considered were Variety and Biofertilizers. All these varieties were obtained from National Grain Legumes Research Program, Khajura and biofertilizers of Agricare Nepal Pvt. Ltd. Company was used in the experiment. The size of plot receiving each treatment was 2mx3m. The distance between each plot was 0.5m whereas the distance between each replication was 1m. For the treatments, seeds were soaked for 8 hours and treated with Rhizobium @10g per kg seed two hour before sowing whereas Trichoderma and pseudomonas were drenched in soil @ 10 ml/liter water one day before sowing.

Agronomic practices

The seeds were soaked for 8 hours in water before to sowing. FYM was applied @6t/ha one month before sowing whereas chemical fertilizers were applied @ 20:20:20 Kg NPK/ha just before sowing. On April 3rd, two seeds per spot were dibbled at spacing of 10cm in lines made 30 cm apart in all the 54 plots. Thinning was done 10 days after sowing to maintain spacing of 30cmx10cm. Irrigation was done during the time of pod formation. Manual weeding was done 30 days after sowing. The crop was harvested manually at two times i.e. 60 DAS and 70 DAS.

Observation and data collection

During the study, all the morphological characteristics like plant height, root length, number of effective and non-effective root nodules per plants were taken from randomly selected 5 plants at 45 and 60 days of sowing. For the measurement of underground portion, five randomly selected plants were uprooted from each plot from the destructive row.

Plant height

The plant height was taken from randomly selected 10 plants from different rows except border rows and was averaged for each treatment. The plant height, measured in centimeter, was taken from the base of the plant to the tip of the leaf.

Root length

Root length (cm) was measured from upper surface of the soil to the tip of the root, after uprooting the plants from each treatment. The average value of sampled plants was taken as root length.

Shoot root ratio

The shoot-root ratio was calculated by dividing the length of the shoot (aboveground portion) by the length of the root (below-ground portion) of a plant.

Shoot Root Ratio (SRR) = $\frac{\text{Shoot Length}}{\text{Root Length}}$

Root branches

Root branches was measured by counting all the secondary lateral root branches from the uprooted plants from destructive rows of each treatment.

Root nodules

All the root nodules were counted from 5 uprooted plants, and averaged to count the total number of root nodules. Root nodules were then pressed in white sheet to observe the color. The round and large nodules producing pink or reddish color, when pressed were considered as effective nodules and those producing green color and lack pink or reddish color, irregular shape smaller in size were considered as non-effective nodules.

Days to flowering

Days to flowering was counted when 50 % of the plant showed flowers and started to form pod.

Leaf area index (LAI)

LAI was taken from randomly selected 10 plants from each treatment. At flowering stage, 3 leaves from each plant i.e. small, medium and large were taken for measuring leaf area by using YMJ-D Portable Leaf Area Meter. The average mean leaf area was calculated and using formula LAI is measured.

 $Leaf Area Index = \frac{Leaf Area}{Ground Area}$

Pod length

The length of pod was measured in centimeter from top to bottom of the pod, using scale for randomly selected 20 pods from each treatment. The values were then averaged for pod length.

Grains per pod

After harvesting, 20 pods were randomly selected for counting the number of grains in each pod. The average value was taken as seed per pod for each treatment.

Grain yield

The total grains obtained from each treatment after threshing was taken for grain yield. For this, harvesting was done for $1m^2$ land area and then calculated in tons per hectare.

Data analysis

The data were subjected to analysis of variance, after recording all the growth parameters, yield attributing traits and yield for each treatment. Least significance difference test was done to compare the treatment means at 5% level of significance. For conducting these tests, the software used was R-studio 4.2.3.

RESULTS & DISCUSSION

Root parameters

Among the mung bean varieties, SML-668 was statistically superior in generating the number of root branches but was inferior in yielding total number of nodules and effective nodules. Kalyan variety was found to be quick in generating number of root branches, total number of nodules and effective nodules per plant as observed in Table 2. Similarly, Pratigya variety of mung bean, having statistically lowest number of root branches was quite good in yielding number of nodules and effective nodules per plant (p < 0.05). The results revealed that the number of nodules per plant was inversely proportional to the number of root branches at 60 DAS.

Within the biofertilizers compared in this study, the combination RTP showed statistically highest value for all the root parameters at 45 DAS and 60 DAS followed by RP and RT respectively. The treatment without application of any biofertilizers (control) showed the lowest value for root branches, nodules and effective nodules per plant. Rhizobium when combined with pseudomonas or trichoderma performed better in generating root parameters than when applied alone (R). This result is coherent with the findings by Sah et al., (2021), who reported proper nodule formation due to increased availability of phosphorous by Pseudomonas. Likewise, the combination of Trichoderma and Pseudomonas (TP) could not perform better in root parameters in absence of Rhizobium, indicating its importance in leguminous crop like mung bean. Also, the result showed higher percentage of ineffective nodules (14.9%) at 45 days after sowing (DAS) than at 60 DAS (11.9%) explaining increase in effective nodules at the time of flowering.

Treatment	Total root branches per plant (45 DAS)	Total root branches per plant (60 DAS)	Total nodules per plant (45 DAS)	Total nodules per plant (60 DAS)	Effective nodules per plant (45 DAS)	Effective nodules per plant (60 DAS)
Varieties						
Kalyan	16.08ª	18.65 ^b	27.33ª	33.16 ^{ab}	23.5ª	29.33ª
Pratigya	15.53 ^b	18.70 ^b	27.05ª	33.22ª	22.66 ^{ab}	29.38ª
SML-668	16.23ª	19.77 ^a	25.72 ^b	31.94 ^b	22.00 ^b	27.88 ^b
SEM (±) F-test	0.13 **	0.18 ***	0.33 **	0.43 ns	0.35 *	0.43 *
LSD	0.38	0.52	0.94	-	1.00	1.24
Biofertilizers						
Control	13.04 ^d	16.33°	21.66 ^d	26.66°	17.00 ^d	22.55 ^e
R	16.26 ^{bc}	19.08 ^b	26.77°	32.55 ^{cd}	23.11 ^{bc}	28.88 ^{cd}
RT	16.55 ^b	19.62 ^b	27.33 ^{bc}	33.88 ^{bc}	23.55 ^{bc}	30.11 ^{bc}
RP	16.73 ^{ab}	19.31 ^b	28.22 ^b	34.88 ^b	24.44 ^{ab}	31.11 ^{ab}
RTP	17.17ª	20.84ª	29.88ª	36.88ª	25.55ª	32.44 ^a
ТР	16.94°	19.06 ^b	26.33°	31.77 ^d	22.66°	28.11 ^d
SEM (±)	0.19	0.25	0.46	0.61	0.49	0.62
F-test	***	***	***	***	***	***
LSD	0.54	0.73	1.34	1.76	1.42	1.76
A*B	NS	NS	NS	NS	NS	NS
CV	3.58	4.03	5.25	5.62	6.54	6.38
Grand mean	15.95	19.04	26.7	32.77	22.72	28.87

Table 1. Effect of varieties and biofertilizers on root parameters of mung bean at Tikapur, Kailali, 2022

LSD: least significant difference; CV: coefficient of variance; SEM: standard errors of Means; *: significant at ≤ 0.05 level of significance; **: significant at ≤ 0.01 ; ***: significant at ≤ 0.001 level of significance; NS: Non-Significant

Days to flowering

The results showed that neither the varieties nor the biofertilizers exerted significant effect on 'days to flowering' of mung bean. However, flowering delayed by nearly one

day in SML-668 taking 36.72 days when compared with Kalyan, which took almost 35.88 days to flower. Regarding the biofertilizers, all the combinations containing trichoderma showed slight delay in flowering, whereas rhizobium applied field was quick to show up flowers than the control. Zaman et al. (2011) reported early flowering in inoculated plants when compared with control in chickpea. The combination RT was relatively late to show up the flowers followed by RTP, TP and RP in this study. Several reports indicated early flowering when treated with biofertilizers like Azotobactor, Phosphate solubilizing bacteria, Rhizobium, Azospirillum and Vesicular-arbuscular mycorrhizae and rhizobium in French bean (Meena et al., 2018; Ndlovu, 2015).

Leaf area index

The variety SML-668 showed statistically highest value for leaves are index followed by Pratigya and Kalyan respectively, when measured at flowering time. Kalyan showed 5.55 % and 14.06% lower value of leaf area than Pratigya and SML-668 respectively. Similarly, the leaf area index at 60 DAS was found to be the highest with the combination RTP, and the lowest in the control treatment. The LAI obtained from combinations of Rhizobium with other biofertilizers were statistically higher than Rhizobium, when applied alone. Statistically R and TP were at par in yielding LAI, but showed the lowest value among the biofertilizer treatments in this study. In line with our findings, increased LAI due to increased number of microbes in soil as a result of biofertilizers' application has also been reported by Kaur and Kumar (2020).

Treatment	Days to flowering	LAI at flowering	Shoot root ratio (45 DAS)	Shoot root ratio (60 DAS)
Varieties				
Kalyan	35.88 ^b	3.91°	3.85°	3.70°
Pratigya	36.16 ^{ab}	4.14 ^b	4.01 ^b	3.86 ^b
SML-668	36.72ª	4.55ª	4.28 ^a	4.10 ^a
SEM (±)	0.26	0.06	0.03	0.02
F-test	ns	***	***	***
LSD	-	0.18	0.08	0.07
Biofertilizers				
Control	35.77 ^b	3.44 ^d	4.40^{a}	4.15 ^a
R	35.66 ^b	3.99°	4.07 ^b	3.92 ^b

Table 2. Effect of varieties and biofertilizers on days to flowering, leaf area index, shoot root ratio of mung bean at Tikapur, Kailali, 2022

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RT	37.00 ^a	4.36 ^b	3.99 ^{bc}	3.89 ^{bc}	
RP	36.22 ^{ab}	4.44 ^b	3.94°	3.81°	
RTP	36.55 ^{ab}	4.90ª	3.80 ^d	3.66 ^d	
TP	36.33 ^{ab}	4.07°	4.05 ^{bc}	3.90 ^{bc}	
SEM (±)	0.37	0.09	0.04	0.03	
F-test	NS	***	***	***	
LSD	-	0.25	0.12	0.1	
A*B	NS	NS	NS	NS	
CV	3.09	6.44	3.26	2.84	
Grand mean	36.25	4.2	4.04	3.89	

LSD: least significant difference; CV: coefficient of variance; SEM: standard errors of Means; *: significant at ≤ 0.05 level of significance; **: significant at ≤ 0.01 ; ***: significant at ≤ 0.001 level of significance; NS: Non-Significant

Shoot root ratio

Both the factors i.e. varieties and biofertilizers had significant influence on shoot root ratio as observed in Table 2, SML-668 showed vigorous growth and hence had significantly highest shoot root ratio (lengths) followed by Pratigya and Kalyan respectively. The result showed nearly four times (3.66 to 4.15) increase in shoot length as compared to root length in mung bean, as evidenced in Table 2. In the same way, the control plot exhibited significantly highest shoot root ratio over all other treatments containing biofertilizers, revealing relatively shallow growth of root in those plots. The combination RTP exhibited lowest shoot root ratio followed by RP showing the influence of biofertilizers in increasing the length of root. At the same time, this data also explains the substantial growth of shoot with increase in the length of root.

Length of pod

Statistically significant differences in pod lengths were observed among the varieties of mung bean as shown in Table 3. The variety SML-668 had significantly longest pod size (88.53mm) followed by Pratigya (82.3 mm) and Kalyan (78.73 mm). Neupane et al., (2023) reported that Pratigya outperformed Kalyan in pod length. The vigorous growth habit of SML-668 revealed by the number of root branches, leaf area index and shoot root ratio contributed to the increased length of pod in this variety, as compared to other varieties. Kalyan showed significantly smallest pod size attributed by

its low LAI, shoot root ratio despite having the higher number of effective nodules than

There was no significant difference among the biofertilizers in the length of pods at 5% level of significance. Numerically the treatment RP exhibited the longest length of pod whereas smallest pod was observed in the control.

Number of grains per pod

SML-668.

Despite the significant differences in the length of pod between SML-668 and Kalyan, the number of grains per pod were statistically at par but were superior to Pratigya variety. Early flowering might have led to higher number of grains per pod despite its poor growth habit and length of pod in Kalyan over Pratigya variety. Statistically higher number of grains per pod in bio fertilizer treatments over control showed the influence of biofertilizers in yielding total number of grains per pod. The control plot yielded only 9.66 grains per pod which was 10.30%, 11.61 %, 10.55%, 11.86 % and 9.88% lesser than R, RT, RP, RTP and TP respectively. Increased number of grains per pod due to use of biofertilizers has also been reported by (Bam et al., 2022; Khanna et al., 2019; Mohammadzadeh et al., 2022).

Treatment Pod length (mm)		Number of grains per pod	Grain yield (kg/ha)
Varieties			
Kalyan	78.73°	10.59ª	1583.88°
Pratigya	82.30 ^b	10.48 ^b	1726.66 ^b
SML-668	88.53ª	10.85ª	1812.77ª
SEM (±) F-test	0.75 ***	0.84	8.54 ***
LSD	2.16	0.24	24.55
Biofertilizers			
Control	81.25 ^b	9.66 ^b	1581.11°
R	83.12 ^{ab}	10.77^{a}	1712.22 ^ь
RT	83.14 ^{ab}	10.93ª	1733.33 ^{ab}
RP	84.98 ^a	10.80^{a}	1742.22 ^{ab}
RTP	83.68 ^{ab}	10.96^{a}	1765.55ª

Table 3. Effect of varieties and biofertilizers on pod length, number of grains per pod and grain yield of mung bean at Tikapur, Kailali, 2022

ТР	83.02 ^{ab}	10.72ª	1712.22 ^ь
SEM (±)	1.06	0.11	12.08
F-test	ns	***	***
LSD	-	0.33	34.73
A*B	ns	ns	ns
CV	3.83	3.32	2.12
Grand mean	83.2	10.64	1707.77

LSD: least significant difference; CV: coefficient of variance; SEM: standard errors of Means; *: significant at ≤ 0.05 level of significance; **: significant at ≤ 0.01 ; ***: significant at ≤ 0.001 level of significance; NS: Non-Significant

Grain yield

The result showed significant increase in grain yield in SML-668 over Pratigya and Kalyan varieties. Statistically lowest amount of grain yield was obtained from Kalyan variety, which was 8.27 % and 12.62% lower than Pratigya and SML-668 respectively. Profuse growth of both the root and shoot parameters improve the grain yield of legumes (Asad et al., 2004; Bam et al., 2022; Itelima et al., 2018). The lowest grain yield from Kalyan variety might be attributed to the lowest number of root branches, leaf area index and shoot root ratio. The higher grain yield from SML-668 over other varieties of mung bean has also been documented by Baraki et al. (2020) in this study.

Statistically, significant improvement in grain yield due to combination of fertilizers was observed in the study with the highest from RTP and the lowest from the control. An increase in grain yield by 11.66 % over control was recorded in RTP. Similarly, the increase of 3.11 %, 1.75 % and 1.23 % increase in grain yield was obtained from RTP, RP and RT over TP. The result revealed that the bio fertilizer combinations containing rhizobium was statistically superior over those without rhizobium. An increase in grain yield by 1.75 % and 1.23 % were observed in RP and RT respectively over Rhizobium (R). The drastic increase in grain yield from biofertilizers applied plots over the control can be attributed to increase in root branches, nodulation resulting in greater nutrient and water availability leading to proper growth, flowering, fruiting and pod formation. This has also been reported in several studies (Bam et al., 2022; Chaudhary, 2010; Singh & Pareek, 2003).

CONCLUSION

Among the varieties studied, the pipeline variety SML-668 was superior the growth, yield and yield attributes except for nodulation followed by Pratigya. Kalyan

variety performed poorly in Tikapur condition. Thus SML-668 and Pratigya variety could be recommended in similar cropping system for better yield. Similarly, application of Rhizobium in combination with either Trichoderma or Pseudomonas showed better growth and yield of mung bean, than when applied alone. Hence rhizobium inoculation is highly recommended for successful production of mung bean. However, further study on SML-668 at both the lowland and upland conditions in rice-wheat cropping system need is suggested for comprehending the finding of this research.

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