

# Effects of different levels of phosphorus and *bradyrhizobium* inocula on the productivity and protein content of mungbean (*vigna radiata* L. wilczek)

## Abstract

Phosphorus (P) is one of the major limiting nutrients to mung bean production. However, the use of imbalance P nutrient and lack of inocula decreased the mungbean yield in Bangladesh. Therefore, this study has been undertaken to study the effect of different phosphorus levels and *Bradyrhizobium* inocula on the yield and protein content of mungbean. Treatment comprised of four levels of phosphorus - P<sub>0</sub>, P<sub>30</sub>, P<sub>60</sub>, P<sub>90</sub> (0, 30, 60 and 90 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> respectively) and strains of *Bradyrhizobium* inocula - I<sub>0</sub>, I<sub>1</sub>, I<sub>2</sub> and I<sub>3</sub> (not inoculated, inoculation with BINA-MB-THA 301, BINA-MB 441 and BINA-MB 301). The highest seed yield (1066.3 kg ha<sup>-1</sup>) was obtained from P<sub>60</sub>, followed by P<sub>90</sub> (957.7 kg ha<sup>-1</sup>) and P<sub>30</sub> (870.5 kg ha<sup>-1</sup>) while the lowest seed yield (578.9 kg ha<sup>-1</sup>) was obtained from the control. Protein content was highest (24.5 %) in P<sub>90</sub>, followed by P<sub>60</sub> (24.4%) and P<sub>30</sub> (24.4 %), and the lowest (24.1 %) was obtained from control. Inoculation of seed increased the yield and yield components, and harvest index. Better performance of the yield components and protein content of seed was found with the inoculants I<sub>1</sub>. The I<sub>1</sub> strain produce the highest seed yield (982.5 kg ha<sup>-1</sup>) while the lowest yield (670.5 kg ha<sup>-1</sup>) was obtained from uninoculated control plot. The highest protein content of mungbean seed (25.3 %) was estimated when I<sub>1</sub> strain of *Bradyrhizobium* was used for seed inoculation. Among the treatment combinations, P<sub>60</sub>I<sub>1</sub> provided better growth and yield performance and increased protein content of mungbean.

**Keywords:** rhizobium strain, inoculation, yield, protein content, orthogonal comparison

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## Introduction

Mungbean (*Vigna radiata* L. Wilczek) is one of the major and short duration pulse crops grown in pre-monsoon season of Bangladesh. It belongs to the family *Leguminosae* and sub family *Papilionaceae*. The crop is commonly known as green gram, golden gram, mung dal, mungbean but most commonly as 'Mung' in Bangladesh. It is an excellent source of easily digestible protein which complements the stable rich diet in the country. Mungbean seed contains 51 % carbohydrate, 26 % protein, 1 % fat, 4 % minerals, 3 % vitamins and 10 % moisture.<sup>1</sup> Mungbean covered an area of 44006.32 hectares with a production of 37054 tonnes in 2019-20.<sup>2</sup> The average yield of mungbean is about 0.842 t ha<sup>-1</sup>, which is very low compared to the potential yield (1.5-1.6 t ha<sup>-1</sup>) of mung bean in Bangladesh.<sup>3</sup> The yield difference between potential and actual yield indicates the wide scope for increasing yield of mungbean in Bangladesh with improved management practices and by using proper doses of fertilizer.

Optimum amount phosphorus supply is important to fix nitrogen in the soil by the legumes. As mungbean is a legume crop, it responds well to added phosphorus.<sup>4</sup> Phosphorus deficiency causes yield reduction by limiting plant growth.<sup>5</sup> It influences nutrient uptake by promoting root growth and nodulation.<sup>6</sup> The use of phosphorus nutrient has steadily increased overtime. The P consumption in 1981 was 80000 ton which in 2016 increased to 210000 ton in Bangladesh.<sup>7</sup> Mungbean responds favourably to phosphorus fertilization.<sup>8</sup> Phosphorus enhance the uptake of nitrogen, thus increase nitrogen content in the crop which increase protein content of mungbean.<sup>9</sup>

Presently, a number of organisms like *Rhizobium/Bradyrhizobium* have been identified to use as biological agent for fix atmospheric nitrogen by symbiotic process with legume crops and make it available to the plants. Bangladesh Institute of Nuclear Agricultural

(BINA) isolated some *Rhizobium/Bradyrhizobium* strains especially for mungbean cultivar. To reduce the production cost and to fulfill the demand, more pulse production could be achieved through seed inoculation with *Rhizobium/Bradyrhizobium*. As per Franco,<sup>10</sup> *Rhizobium/Bradyrhizobium* strains in association with the host plant were able to fix approximately 20 % of the atmospheric nitrogen through the world annually. Singh et al.<sup>11</sup> reported that seed inoculation of mungbean with *Bradyrhizobium* increased the protein content of mungbean. Hence, there is a large scope of utilizing the biological nitrogen fixing technology for obtaining more protein rich food from mungbean and also to improve the nitrogen status of soils by selecting efficient strains of mungbean inocula. The use of effective strain of mungbean inocula can play a vital role in improving yield and soil health. The strains of mungbean inocula identified by BINA which are suitable for better yield performance of mungbean, but the potentialities of different strain of *Bradyrhizobium* may vary depending upon the soils and climatic conditions. Therefore it is urgently needed to verify the potentialities of different strains of *Bradyrhizobium* in mungbean crop. Limited studies are available to find out the effect of phosphorus and inoculum separately, however, to the best of our knowledge, no studies on the combined effect of these two factors on mungbean are available in Bangladesh. Considering the above facts, the present study has been undertaken to find out the optimum level of phosphorus and suitable *Bradyrhizobium* strain to achieve high yield and protein content of mungbean.

## Materials and methods

### Site of the research

The field experiment was conducted at the Agronomy Field Laboratory, Bangladesh Agricultural University (BAU), Mymensingh, during the period of March to June (2013) in the *Kharif-1* season

to study the effect of phosphorus level and different strain of *Bradyrhizobium* inocula on the yield performance and protein content of mungbean (cv. BINA Mung-2). Geographically the experimental field was located at 24.75° north latitude and 90.50° east longitude at an altitude of 18 m above the mean sea level.

### Initial Soil properties at 0-15 cm soil depth

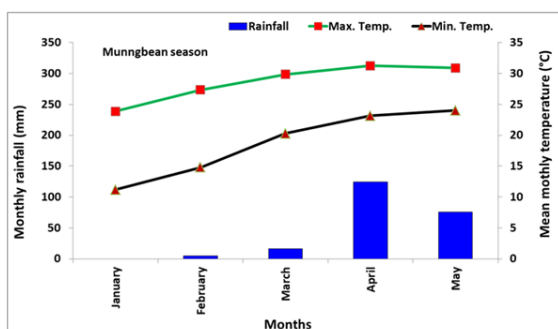
The soil is belongs to the Old Brahmaputra Alluvium Soil under Agro-Ecological Zone 9 (AEZ-9, Old Brahmaputra Floodplain) under Sonatala series.<sup>12</sup> The experimental site falls into Non-calcareous Dark Grey Floodplain soils (Aeric Haplaquept). The land topography was medium high, silty loam soil texture. The soil was sampled from six to seven random spots at 0-15 cm depth of the experimental plot and analyzed for both physical and chemical properties. The initial soil characteristics at 0-15 cm soil depth before laying out the experiment are presented in Table 1.

**Table 1** Soil physico-chemical characteristics at 0-15 cm soil depth before starting the experiment

Soil properties	value
pH	6.5
Electrical Conductivity (dS m <sup>-1</sup> )	0.26
Organic Carbon (%)	0.94
Total N (%)	0.09
Available P (mg kg <sup>-1</sup> )	6.6
Available S (mg kg <sup>-1</sup> )	11.5
Available Zn (mg kg <sup>-1</sup> )	0.8
Available Fe (mg kg <sup>-1</sup> )	55.4
Available B (mg kg <sup>-1</sup> )	0.3
Exchangeable K (me %)	0.07
Textural class	Silt loam
Sand (%)	60
Silt (%)	18
Clay (%)	22
Bulk density (g cm <sup>-3</sup> )	1.35

### Weather condition during study period

The experimental site is under a subtropical monsoon climatic region with a mean monthly maximum and minimum temperature of 29 °C and 19 °C, respectively, with total rainfall of 217 mm during the mungbean growing season (Figure 1). During the mungbean growing seasons (March-May), the highest rainfall of 125 mm was received in April, followed by 76 mm in 2011, while only 5.1 mm and 21.4 mm of rainfall was received in February and March, respectively in 2013. Most of the rainfall was received in April, accounting for nearly 44 % of the total rainfall.



**Figure 1** Mean monthly temperature and total rainfall during mungbean growing season of the research area, recorded from the local weather station.

### Treatments and experimental design

BINA Mung-2 was used as the test crop. The cultivar is suitable for growing in the summer season.<sup>13</sup> Four levels of Phosphorus @ 0, 30, 60 and 90 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> form TSP were applied and four inocula namely not inoculated (I<sub>0</sub>), Inoculation with BINA-MB-THA 301 (I<sub>1</sub>), Inoculation with BINA-MB 441 (I<sub>2</sub>), Inoculation with BINA-MB 301 (I<sub>3</sub>) were used in the experiment. Fresh seeds of mungbean were dipped in the culture broth for 18-20 minutes. The experiment was laid out in a Randomized Complete Block Design (RCBD) having three replications. The unit plot size was 4.0 m x 3.0 m.

### Crop management

Mungbean seeds were sown in the furrow on 7<sup>th</sup> March, 2013. The line to line distance was 30 cm with continuous sowing in the line. Other than phosphorus, the crop was fertilized during final land preparation at the rate of 20 kg N ha<sup>-1</sup> as urea, 20 kg K ha<sup>-1</sup> as muriate of potash (MP), 10.8 kg S ha<sup>-1</sup> as gypsum and 1 kg B ha<sup>-1</sup> as boric acid, as recommended by the Pulses Research Centre, Bangladesh Agricultural Research Institute. All fertilizers were applied by broadcasting and were mixed with soil thoroughly. Weeding and thinning were done at 18<sup>th</sup> and 35<sup>th</sup> days after sowing (DAS). Plant to plant distance was maintained at 6-7 cm. During the experimental period, there was heavy rainfall for several times. Hence, irrigation was not necessary but it was essential to drainage the excess water from the field. Jute hairy caterpillar and pod borer were successfully controlled by the application of Malathion 57 EC @ 1.5 L ha<sup>-1</sup> at the time of 50 % pod formation stage (55 DAS). At the time when 80 % of the pods turned brown colour, the crop was assessed to attain maturity. The crop was harvested on 21<sup>st</sup> May, 2013 from pre-selected 4.0 m<sup>2</sup> area.

### Data and their estimation procedures

Yield components for mungbean, i.e. plant height (cm), branches plant<sup>-1</sup>, pods plant<sup>-1</sup>, sterile pods plant<sup>-1</sup>, aborted ovule pod<sup>-1</sup>, pod length (cm), seeds pod<sup>-1</sup>, dry weight plant<sup>-1</sup> (g), 1000-seed weight, seed and stover yield were recorded using 1 m<sup>2</sup> quadrat from three places in each plot. The pods of each plot were harvested by hand picking as per maturity. Data on yield components were collected from five randomly selected plants plot<sup>-1</sup>. Before harvesting, five plants were selected and uprooted randomly from each of the plot for data recording. Harvest index was calculated with the help of following formula:

$$\text{Harvest index (HI \%)} = \frac{\text{Seed yield}}{\text{Biological yield}} \times 100$$

Seed yield was recorded at 11 % moisture content. Stover weight was determined after oven-drying at 70 °C to a constant weight. Seed and stover yields were taken from the central part of each plot after discarding the border rows. The collected data were compiled and tabulated properly for statistical analysis. Protein content of mungbean seed was estimated by the standard micro Kjeldahl method (total nitrogen estimation method).<sup>14</sup>

### Statistical analysis

The collected data were compiled and analyzed statistically using the analysis of variance (ANOVA) technique with the help of MSTAT-C programme and the mean separation were adjudged by Duncan's Multiple Range Test.<sup>15</sup>

## Results and discussion

### Effect of phosphorus on growth and yield of mungbean

Different phosphorus levels had significant effect on plant characters, yield and yield attributes of mungbean (Table 2). Plant treated with P<sub>60</sub> gave the tallest plant (70.0 cm) and it was statistically identical with P<sub>30</sub> (67.8 cm) and P<sub>90</sub> (67.1 cm). Plant grown without

phosphorus (control) produced the shortest plant (64.4 cm) though it was identical to the plant when received P<sub>90</sub> (67.1 cm). These findings are collaborate with the results reported by Patel and Patel<sup>16</sup> and Imran et al.<sup>17</sup> where they observed that plant height of mungbean showed superiority at 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> followed by 40 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>. The highest number of branches plant<sup>-1</sup> (2.6) was counted from the treatment P<sub>60</sub> which was statistically similar to P<sub>90</sub> (2.5) and P<sub>30</sub> (2.4). The lowest number of branches plant<sup>-1</sup> (1.8) was counted from control.

**Table 2** Effects of phosphorus level on various plant characters, yield and yield attributes of mungbean

Level of phosphorus (kg/ha P <sub>2</sub> O <sub>5</sub> )	Plant height (cm)	Branches/ plant	Pods/ plant	Sterile pods/ plant	Aborted ovule/pod	Pod length (cm)	Seeds/ pod	Dry weight/ plant (g)
P0	64.4b*	1.8b	10.5c	2.1a	1.8a	4.8d	8.4c	10.4d
P30	67.8a	2.4a	13.2b	1.9b	1.3c	5.6b	9.2b	11.2c
P60	69.9a	2.6a	15.0a	1.4c	1.0d	6.1a	9.8a	12.3a
P90	67.1ab	2.5a	14.2ab	2.0ab	1.4b	5.3c	9.5ab	11.8b
CV (%)	5.4	13.2	5.7	8.4	6	3.7	5.4	4.5

\*In a column, means followed by the different letters differ significantly at 1% levels of probability;

P<sub>0</sub> = No phosphorus, P<sub>30</sub> – 30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, P<sub>60</sub> – 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, P<sub>90</sub> – 90 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>

The results are in agreement with the findings of Singh *et al.*<sup>6</sup> where they reported that number of branches plant<sup>-1</sup> of mungbean increased with increased level of phosphorus up to 26.4 kg P ha<sup>-1</sup>. Different levels of phosphorus significantly affected the number of pods plant<sup>-1</sup> (Table 3). Highest number of pods plant<sup>-1</sup> (15.0) was recorded from P<sub>60</sub> treatment. From the study it was observed that P<sub>30</sub> and P<sub>90</sub> gave the identical number of pods plant<sup>-1</sup> but statistically lower than P<sub>60</sub>. The lowest number of pods plant<sup>-1</sup> (10.5) produced from control treatment (P<sub>0</sub>). The results suggested that the number of pods plant<sup>-1</sup> increases with the increased level of phosphorus up to P<sub>60</sub>, and afterwards it started to decline. These findings are in close conformity with the findings of Rao *et al.*<sup>18,19</sup> where they reported that number of pods plant<sup>-1</sup> increased significantly with increasing phosphorus levels from 0 to 50 kg ha<sup>-1</sup>. The plants of control plot (that received no phosphorus fertilizer) produced significantly the highest number (2.1) of sterile pods plant<sup>-1</sup>. It was statistically similar with high doses of phosphorus fertilizer (P<sub>90</sub>). However, P<sub>90</sub> and P<sub>30</sub> produced statistically identical number of sterile pods plant<sup>-1</sup>. On the other hand, the lowest number of sterile pods plant<sup>-1</sup> was observed from P<sub>60</sub> treatment. The result indicated that both higher and lower doses of phosphorus beyond P<sub>60</sub> enhanced the production of more sterile pods plant<sup>-1</sup>. The highest number of aborted ovules pod<sup>-1</sup> (1.8) was recorded from control (P<sub>0</sub>) treatment which differed significantly from other phosphorus levels. The lack of phosphorus nutrients might

be affected the phenological development of the crop and increased the rate of abortion, which led to produce the maximum number of aborted ovules pod<sup>-1</sup>. The lowest number of aborted ovules pod<sup>-1</sup> (1.0) was recorded from P<sub>60</sub>. The result indicated that P<sub>60</sub> significantly reduced the number of aborted ovules pod<sup>-1</sup>. The result of the study further indicated that both lower and higher doses of phosphorus increased abortion of ovules pod<sup>-1</sup>. Plant received P<sub>60</sub> significantly increased the pod length (6.1 cm) which was followed by P<sub>30</sub> (5.6 cm) and P<sub>90</sub> (5.3 cm). The shortest pod length (4.76 cm) was measured from the control (P<sub>0</sub>) plot. A highly significant variation was observed among phosphorus levels in respects of the number of seeds pod<sup>-1</sup>. It was observed that P<sub>60</sub> produced the highest number of seeds pod<sup>-1</sup> (9.8) which was statistically identical to the crop treated with P<sub>90</sub> (9.5). The lowest number of seeds pod<sup>-1</sup> (8.4) was counted from the control plot. This might be due to the insufficiency of phosphorus nutrient, which lowers the uptake of other nutrients and hampers the growth and development, flowering and pod setting of the crop and resulted in lower number of seeds pod<sup>-1</sup> in control (P<sub>0</sub>) plot. These findings are closely related with the findings of Patel and Patel<sup>16</sup> who found that the number of seeds pod<sup>-1</sup> increased significantly with increasing levels of phosphorus from 0 kg ha<sup>-1</sup> to 60 kg ha<sup>-1</sup>. Dry weight plant<sup>-1</sup> significant affected due to different levels of phosphorus. Maximum dry weight plant<sup>-1</sup> (12.27 g) was found from P<sub>60</sub> while the lowest dry weight plant<sup>-1</sup> (10.42 g) was obtained from the control plot.

**Table 3** Effects of Bradyrhizobium strain on various plant characters, yield and yield attributes of mungbean

Bradyrhizobium strain	Plant height (cm)	Branches/ plant	Pods/ plant	Sterile pods/ plant	Aborted ovules/pod	Pod length (cm)	Seeds/ pod
I0	64b*	2.0b	11.5c	2.4a	2.0a	5.0c	8.4b
I1	70 a	2.7a	15.5a	1.5d	0.9d	5.7a	9.6a
I2	68 a	2.5a	12.8b	1.7c	1.2c	5.5b	9.6a
I3	67a	2.2b	11.9bc	1.8b	1.3b	5.5b	9.4a
CV (%)	5.4	13.2	8.39	8.4	6	3.7	5.4

I<sub>0</sub> – Not inoculated; I<sub>1</sub> – Inoculation with BINA-MB-THA 301; I<sub>2</sub> – Inoculation with BINA-MB 441 and I<sub>3</sub> – Inoculation with BINA-MB 301

\* In a column, means followed by the different letters differ significantly at 5 % levels of probability

The highest 1000-seed weight (25.2 g) was recorded from P<sub>60</sub> treatment which was followed by P<sub>90</sub> (23.6 g) (Figure 2). The crop was fertilized with P<sub>90</sub>, P<sub>30</sub> and control gave statistically identical

results. These findings are partly similar with the findings of Reddy *et al.*<sup>20</sup> where they observed that application of 50 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> resulted highest 1000-seed weight of mungbean. The seed yield was

significantly influenced by different phosphorus levels (Figure 2). It was found that  $P_{60}$  produced the highest seed yield (1066 kg ha<sup>-1</sup>) while the lowest seed yield (578.9 kg ha<sup>-1</sup>) was obtained from the control. Seed yield increased due to the application of  $P_{60}$  fertilizer, it might have fulfilled the requirements of the crop which helped better uptake of other plant nutrient at balance proportion to result in better plant growth and subsequently improve the yield contribution attributes and finally increased the seed yield. Significantly the highest stover yield m<sup>-2</sup> (476.7 g) was produced by the crop when received  $P_{60}$ . The lowest amount of stover yield (3576 kg) obtained from control plot (Figure 2). The highest plant height, number of branches plant<sup>-1</sup>, number of pods plant<sup>-1</sup>, number of leaves plant<sup>-1</sup> might be contributed to increase stover yield of mung bean. Similar finding was found by

Sharma et al.<sup>21</sup> reported that stover yield of mungbean increased with increase of phosphorus up to or equivalent of 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>. Different phosphorus levels exerted a significant influence on the harvest index of mungbean (Figure 2). Harvest index was recorded maximum (22.7 %) with  $P_{60}$ . The harvest index was statistically similar under  $P_{90}$  and  $P_{30}$  treatments. The lowest harvest index (15.8 %) was obtained from the control plot. Response curve in orthogonal comparison in regression indicates that the optimum phosphorus level for mungbean cultivation was 63 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and its corresponding seed yield should be 1307.0 kg ha<sup>-1</sup> and seed yield would be reduced by 0.98 kg ha<sup>-1</sup> for reducing 1 kg of phosphorus fertilizer (Figure 3). These findings are similar with the findings of Raundal et al.<sup>22</sup> where they reported that application of 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> significantly increased the seed yield of mungbean.

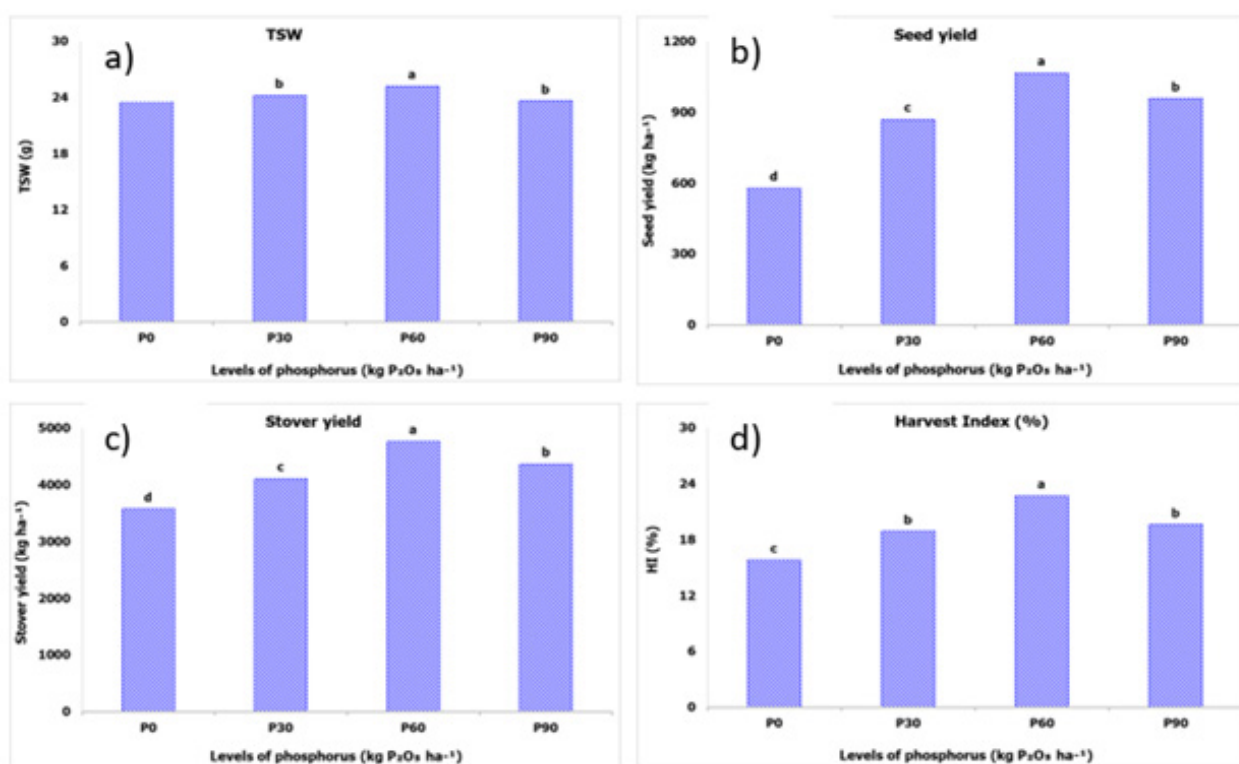


Figure 2 Effects of phosphorus level on thousand seed weight – TSW (a), seed yield (b), stover yield (c) and harvest index (%) (d) of mungbean.

$P_0$  = No phosphorus,  $P_{30}$  – 30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>,  $P_{60}$  – 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>,  $P_{90}$  – 90 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>

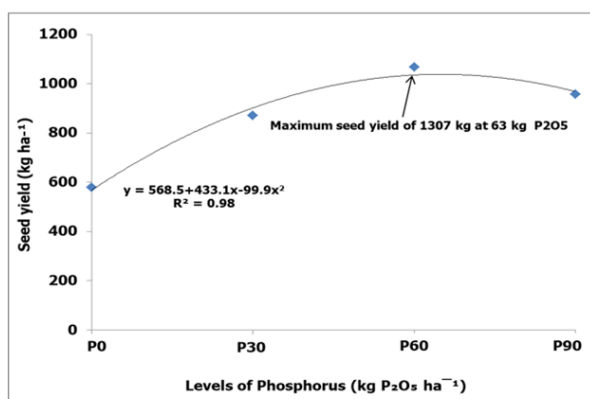


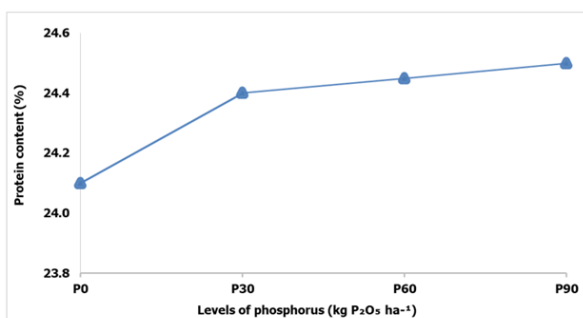
Figure 3 Response curve showing the relationship between level of phosphorus and seed yield of mungbean;  $P_0$  = No phosphorus,  $P_{30}$  = 30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>,  $P_{60}$  = 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and  $P_{90}$  = 90kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>.

The estimated protein content of Mungbean seed varied significantly due to different phosphorus levels (Figure 4). The highest protein content of mungbean seed was recorded from  $P_{90}$  treatment. The lowest protein content was estimated from the control plot. The figure indicates that the protein content of mungbean seed increased with the increased phosphorus levels.

### Effect of Bradyrhizobium strain

Different strain of *Bradyrhizobium* had significant effect on plant height, number of branches plant<sup>-1</sup>, number of sterile pods plant<sup>-1</sup>, number of aborted ovule pod<sup>-1</sup>, pods number plant<sup>-1</sup>, Pod length, number of seeds pod<sup>-1</sup>, dry weight plant<sup>-1</sup>, stover yield, seed yield kg ha<sup>-1</sup>, thousand seed weight and harvest index (Table 2). The tallest plant (70 cm) was obtained when crop was treated with  $I_1$  strain which was statistically identical to  $I_2$  (68 cm) and  $I_3$  (67 cm). The shortest plant height (64 cm) was obtained from the crop when the seed were uninoculated. These might be due to continuous nitrogen supply by *Bradyrhizobium* inoculum to the plant after a stage of growth and

ultimately enhanced plant height. These results are close related with the findings of Kavathiya and Pandey<sup>23</sup> stated that inoculated seed gave significantly increased plant height compared with uninoculated control. Significantly the maximum number of branches plant<sup>-1</sup> (2.7) was found when the seeds were inoculated with I<sub>1</sub> strain which was statistically similar with I<sub>2</sub> (2.5). The lowest branches number plant<sup>-1</sup> was noticed from the control plot (2.0), though it was statistically identical to the crop treated with I<sub>3</sub> (2.2). The plot treated with I<sub>1</sub> produced the maximum number of pods plant<sup>-1</sup> (15.5). The inoculum strain I<sub>2</sub> and I<sub>3</sub> produced statistically identical number of pods plant<sup>-1</sup> 12.8 and 11.9 respectively, where I<sub>3</sub> and I<sub>0</sub> gave statistically similar result and the lowest pod number plant<sup>-1</sup> (11.5) was observed from control (I<sub>0</sub>) strain.

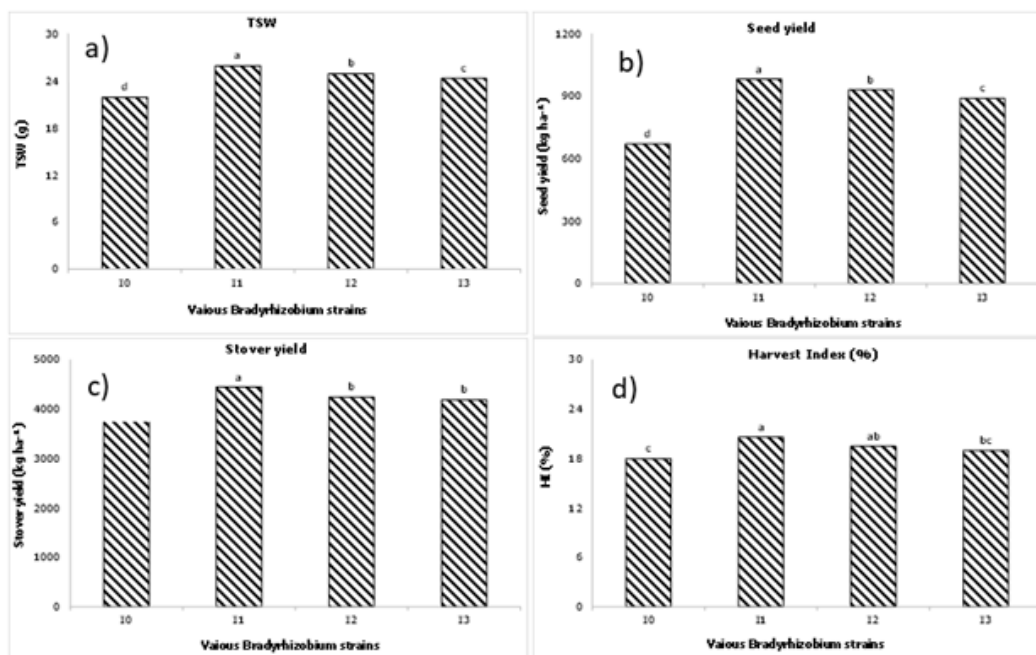


**Figure 4** Protein content of mungbean as influenced by phosphorus levels; P<sub>0</sub> = No phosphorus, P<sub>30</sub> = 30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, P<sub>60</sub> = 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and P<sub>90</sub> = 90 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>.

The highest number of sterile pods plant<sup>-1</sup> (2.4) was found from the control plot (I<sub>0</sub>) followed by inoculation of seed with I<sub>3</sub> and I<sub>2</sub> strain. The lowest number of sterile pods plant<sup>-1</sup> (1.46) was obtained when the crop was treated with I<sub>1</sub> strain of *Bradyrhizobium*. The highest number of aborted ovules pod<sup>-1</sup> (2.0) was counted from the control plot (I<sub>0</sub>) and the lowest number of aborted ovules pod<sup>-1</sup> (0.9) was counted from the plot treated with I<sub>1</sub> strain of *Bradyrhizobium*. This

might be due to the highest potential of I<sub>1</sub> for more nitrogen fixation which enhanced more uptakes of other nutrients and thus reduced the sterility of the crop and produced lower number of aborted ovules. Different strain of *Bradyrhizobium* showed a significant effect on the pod length of mungbean. The highest pod length (5.7 cm) was measured when the seed was treated with I<sub>1</sub> strain of the inoculum. The crop treated with I<sub>2</sub> and I<sub>3</sub> strain produced statistically identical pod length. The lowest pod length (5.0 cm) was measured from the control (I<sub>0</sub>) treatment. The crop treated with I<sub>1</sub> strain produced the highest number of seeds pod<sup>-1</sup> (10.0) which was statistically similar with I<sub>2</sub> (10.0) and I<sub>3</sub> (9.4). The lowest number of seeds pod<sup>-1</sup> (8.4) was counted from control treatment. The result indicated that inoculated plant produced higher number of seeds pod<sup>-1</sup> and increased the pod length than that of uninoculated plant. The lowest dry weight plant<sup>-1</sup> (10.2 g) was obtained from the control plot.

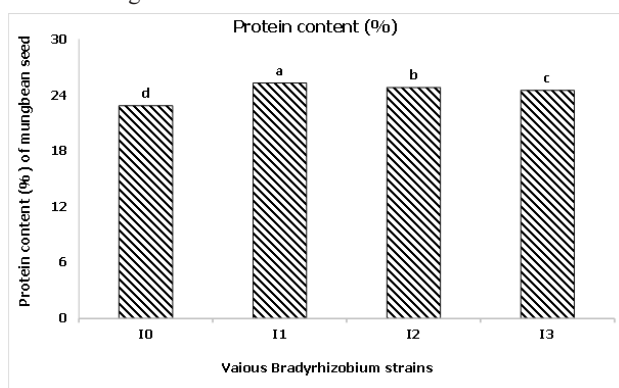
Inoculated and uninoculated plants showed significant variation on the 1000-seed weight of mungbean (Figure 5). The highest 1000-seed weight (25.94 g) was measured when the seed was treated with I<sub>1</sub> strain of *Bradyrhizobium*. The lowest 1000-seed weight (22.0 g) was observed from uninoculated treatment (control). Seed yield of mungbean was significantly the highest (982.50 kg ha<sup>-1</sup>) when I<sub>1</sub> strain of *Bradyrhizobium* was used for seed inoculation and the lowest (670.50 kg ha<sup>-1</sup>) from the control plot (Figure 5). The crop treated with I<sub>1</sub> produced highest amount of stover yield m<sup>-2</sup> (4451 kg ha<sup>-1</sup>) followed by the treatment I<sub>2</sub> and I<sub>3</sub> (Figure 5) But the *Bradyrhizobium* strain I<sub>2</sub> and I<sub>3</sub> produced statistically identical stover yield. These findings are in close conformity with the findings of Chowdhury et al.<sup>24</sup> reported that mungbean seed inoculation with *Bradyrhizobium* strain significantly increased the stover yield by about 50%. Harvest index was also significantly affected by different strain of *Bradyrhizobium* (Figure 5). The highest harvest index (20.58%) was recorded in the crop treated with I<sub>1</sub> strain of *Bradyrhizobium* which was statistically identical with I<sub>2</sub>. The lowest harvest index (18.0 %) was calculated in control plot.



**Figure 5** Effects of Bradyrhizobium strain on thousand seed weight – TSW (a), seed yield (b), stover yield (c) and harvest index (%) (d) of mungbean.

I<sub>0</sub> – Not inoculated; I<sub>1</sub> – Inoculation with BINA-MB-THA 301; I<sub>2</sub> – Inoculation with BINA-MB 441 and I<sub>3</sub> – Inoculation with BINA-MB 301

*Bradyrhizobium* strain played a significant role on protein content of mungbean (Figure 6). It was observed that the maximum protein content (25.28%) was obtained when the seeds were inoculated with I<sub>1</sub> strain of *Bradyrhizobium*, which was followed by I<sub>2</sub> and I<sub>3</sub> strains of *Bradyrhizobium*. The minimum protein content (22.84%) was estimated from the seeds of control plot. This might be due to the absence or minimum number of *Bradyrhizobium* bacteria present in the soil, that could not supply sufficient nitrogen as to the requirements of the crop by the process of biological nitrogen fixation and ultimate low nitrogen content of the seed, thus finally reduced the protein content of mungbean seed.



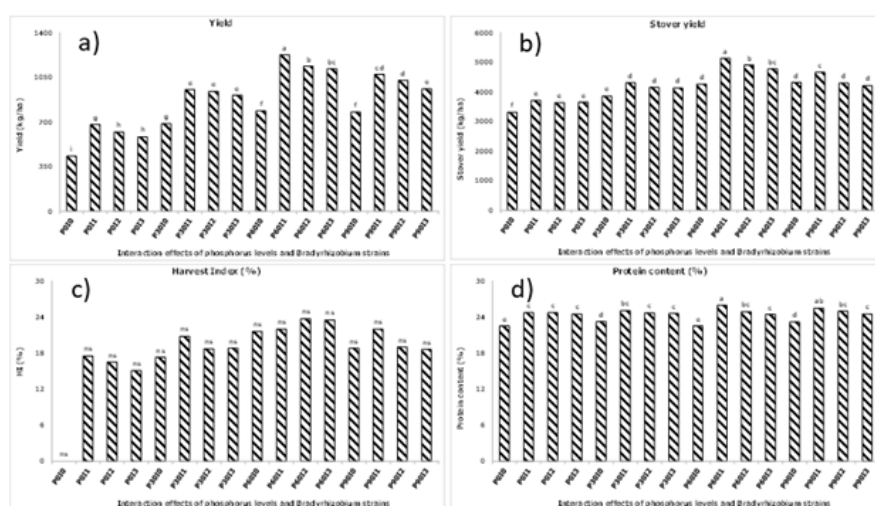
**Figure 6** Effects of *Bradyrhizobium* strain on the protein content of mungbean seed.

I<sub>0</sub> – Not inoculated; I<sub>1</sub> – Inoculation with BINA-MB-THA 301; I<sub>2</sub> – Inoculation with BINA-MB 441 and I<sub>3</sub> – Inoculation with BINA-MB 301

### Interaction effect of different phosphorus level and *Bradyrhizobium* strain

Different phosphorus levels and *Bradyrhizobium* strain interacted to influence sterile pods number plant<sup>-1</sup>, aborted ovules number pod<sup>-1</sup>, seed yield, stover yield and protein content (%) of mungbean but did not significant to influence the plant height, number of branches plant<sup>-1</sup>, pods number plant<sup>-1</sup>, pod length, number of seeds pod<sup>-1</sup>, dry weight plant<sup>-1</sup>, thousand seed weight and harvest index (%) (Table 4 and Figure 7).

The tallest plant (73.2 cm) was found from the plant treated with P<sub>60</sub>I<sub>1</sub>. The shortest plant (58.7 cm) was obtained with P<sub>0</sub>I<sub>0</sub>. This might be due to the lack of phosphorus and nitrogen element of the plant which reduced the cell division, carbohydrate and protein synthesis and also lowers the normal activity of the cambium tissue which results shorter plant height. The maximum number of branches plant<sup>-1</sup> (2.9) was counted when P<sub>60</sub> interacts with I<sub>1</sub> and the lowest number of branches plant<sup>-1</sup> (1.6) was recorded from P<sub>0</sub>I<sub>0</sub>. The highest number of pods plant<sup>-1</sup> (18.5) was obtained from P<sub>60</sub>I<sub>1</sub> and the lowest number of pods plant<sup>-1</sup> was observed with P<sub>0</sub>I<sub>0</sub> (8.3). Significantly the highest number of sterile pods plant<sup>-1</sup> (2.9) was counted in P<sub>0</sub>I<sub>0</sub>. This might be due to the insufficiency of phosphorus and nitrogen nutrients which lowered the plant growth and development, flowering and fruit setting and finally increased the pod sterility of mungbean. The lowest sterile pods number plant<sup>-1</sup> (1.0) was counted from the crop treated with P<sub>60</sub>I<sub>1</sub> which was statistically identical with P<sub>60</sub>I<sub>2</sub>. The highest number of aborted ovules pod<sup>-1</sup> (3.2) was significantly observed from the control plot (P<sub>0</sub>I<sub>0</sub>) and the lowest number of aborted ovules pod<sup>-1</sup> (0.8) was counted when P<sub>60</sub> treated with I<sub>1</sub> and it was statistically identical with P<sub>60</sub>I<sub>2</sub> (0.9) and P<sub>30</sub>I<sub>1</sub> (0.8). The highest pod length (6.3 cm) was obtained by P<sub>60</sub> when coupled with I<sub>1</sub> strain of *Bradyrhizobium* and the shortest pod length (4.4 cm) was recorded from the control (P<sub>0</sub>I<sub>0</sub>) plot. However the highest number of seeds pod<sup>-1</sup> (10.1) was counted when P<sub>60</sub> used coupled with I<sub>1</sub> strain of *Bradyrhizobium*. The lowest number of seeds pod<sup>-1</sup> (7.1) was counted from the control plot where neither phosphorus nor inocula were given. The maximum dry weight plant<sup>-1</sup> (13.8 g) was obtained when P<sub>60</sub> interacts with I<sub>1</sub> strain of *Bradyrhizobium* and the lowest dry weight plant<sup>-1</sup> (8.88 g) was recorded in control plot (P<sub>0</sub>I<sub>0</sub>). Significantly the highest seed yield (1226 kg ha<sup>-1</sup>) was obtained when P<sub>60</sub> was used with I<sub>1</sub> strain of *Bradyrhizobium* and the lowest amount of seed yield (432.3 kg ha<sup>-1</sup>) was found from the control plot (P<sub>0</sub>I<sub>0</sub>) (Figure 7). The highest seed yield was obtained from P<sub>60</sub>I<sub>1</sub> that might be due to the sufficient amount of available phosphorus from P<sub>60</sub> and adequate level of nitrogen supplied by the high potential I<sub>1</sub> strain of *Bradyrhizobium* bacteria by fixing atmospheric nitrogen through the process of biological nitrogen fixation. Significantly the maximum stover yield m<sup>-2</sup> (5129 kg ha<sup>-1</sup>) was obtained when the crop was treated with P<sub>60</sub>I<sub>1</sub> and the lowest straw yield m<sup>-2</sup> (3311 kg ha<sup>-1</sup>) was recorded in control (P<sub>0</sub>I<sub>0</sub>) plot (Figure 7).



**Figure 7** Interaction effects of phosphorus level and different *Bradyrhizobium* strain on yield (a), stover yield (b), harvest index (%) (c) and protein content (%) (d) of mungbean.

P<sub>0</sub> = No phosphorus, P<sub>30</sub> = 30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, P<sub>60</sub> = 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and P<sub>90</sub> = 90 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>

I<sub>0</sub> – Not inoculated; I<sub>1</sub> – Inoculation with BINA-MB-THA 301; I<sub>2</sub> – Inoculation with BINA-MB 441 and I<sub>3</sub> – Inoculation with BINA-MB 301

In case of 1000-seed weight was not significantly affected by phosphorus fertilizer and combined use of *Bradyrhizobium* strain inoculation (Table 4). However, the highest 1000 seed weight (27.7g) was found when P<sub>60</sub> coupled with I<sub>1</sub> strain of *Bradyrhizobium*. The lowest 1000-seed weight (21.7 g) was recorded from control (P<sub>0</sub>I<sub>0</sub>).

Numerically the maximum harvest index (23.7 %) was recorded when P<sub>60</sub> interacts with I<sub>2</sub> strain of *Bradyrhizobium*. The lowest harvest index (14.2 %) was calculated from the absolute control plot (P<sub>0</sub>I<sub>0</sub>) (Figure 7).

**Table 4** Interaction effects of phosphorus level and different Bradyrhizobium strain on various plant characters, yield, yield attributes and protein content of mungbean

Level of phosphorus x Bradyrhizobium strain	Plant height (cm)	Branches/plant	Pods/plant	Sterile pods/plant	Aborted ovules/pod	Pod length (cm)	Seeds/pod	Dry weight/plant (g)	TSW(g)
P <sub>0</sub> I <sub>0</sub>	59	1.6	8.3	2.9a*	3.2a	4.4	7.1	8.9	21.7
P0I1	67	2.1	13.7	1.7fgh	1.0ij	4.9	9	11.9	25.5
P <sub>0</sub> I <sub>2</sub>	65	1.9	10.3	1.8efg	1.4ef	4.8	8.8	10.8	24.9
P <sub>0</sub> I <sub>3</sub>	66	1.7	9.5	1.9def	1.4de	4.8	8.8	10.1	24.3
P <sub>30</sub> I <sub>0</sub>	66	2.1	11.8	2.5b	1.8b	4.9	8.1	10.1	22.3
P <sub>30</sub> I <sub>1</sub>	70	2.7	14.4	1.6gh	0.8k	6	9.3	12.4	25.2
P <sub>30</sub> I <sub>2</sub>	68	2.6	12.6	1.7efgh	1.2g	6	9.7	11.3	24.6
P <sub>30</sub> I <sub>3</sub>	67	2.3	12	2.0de	1.3fg	5.9	9.7	11	22.6
P <sub>60</sub> I <sub>0</sub>	67	2.2	13.7	2.0de	1.3fg	5.6	9.2	11	22.4
P <sub>60</sub> I <sub>1</sub>	73	2.9	18.5	1.0k	0.8k	6.3	10.1	13.8	27.7
P <sub>60</sub> I <sub>2</sub>	70	2.8	14.6	1.1jk	0.9jk	6.2	10	12.6	26.2
P <sub>60</sub> I <sub>3</sub>	70	2.3	13.3	1.3ij	1.1hi	6.2	10	11.8	24.5
P <sub>90</sub> I <sub>0</sub>	62	2.2	12.2	2.4bc	1.7c	5	9.1	10.8	21.1
P <sub>90</sub> I <sub>1</sub>	70	2.9	15.4	1.5hi	1.2gh	5.5	9.9	13	25.4
P <sub>90</sub> I <sub>2</sub>	69	2.7	13.6	2.0de	1.3efg	5.3	9.8	12.1	24
P <sub>3</sub>	67	2.4	12.8	2.1cd	1.5cd	5.3	9.4	11.4	24
CV (%)	5.4	13.2	8.9	8.4	6	3.7	5.4	4.5	2.92

\*In a column, means followed by the different letters differ significantly at 5% levels of probability

P<sub>0</sub> = No phosphorus, P<sub>30</sub> = 30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, P<sub>60</sub> = 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and P<sub>90</sub> = 90 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>

I<sub>0</sub> – Not inoculated; I<sub>1</sub> – Inoculation with BINA-MB-THA 301; I<sub>2</sub> – Inoculation with BINA-MB 441 and I<sub>3</sub> – Inoculation with BINA-MB 301; TSW – Thousand seed weight, HI – Harvest index

Interaction effect of phosphorus and *Bradyrhizobium* strains found significant effect in case of protein content of mungbean (Figure 7). When P<sub>60</sub> used with I<sub>1</sub> strain of *Bradyrhizobium* produced the highest protein content (25.9 %) of mungbean seed which was statistically identical with P<sub>90</sub> at the same strain of *Bradyrhizobium*. Significantly the lowest protein content (22.5 %) was estimated from the seed of absolute control plot.

## Conclusion

The major objective of this study was to evaluate to study the effect of different phosphorus levels and strains of *Bradyrhizobium* inocula on the growth, yield and protein content of mungbean compared with control (not inoculated and no phosphorus levels). In accordance with our hypothesis, the highest seed yield was obtained from 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> while the lowest seed yield was obtained from the control. However, the orthogonal comparison in regression shows that 63 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> is the optimum phosphorus level for the maximization of yield. With increasing phosphorus levels up to 90 kg ha<sup>-1</sup>, the protein content of mungbean seed increased. Inoculation of seed significantly increased the yield components, seed and stover yield, and harvest index of mungbean. The inoculants I<sub>1</sub> (BINA-MB-

THA 301) of *Bradyrhizobium* increased seed yield of mungbean and protein content of mungbean seed. In a nutshell, among the treatment combinations, 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with the inoculants I<sub>1</sub> (BINA-MB-THA 301) of *Bradyrhizobium* improved crop growth and yield, and protein content of mungbean in Bangladesh. Nevertheless, the result of the present study could be verified further experimentation in different agro-ecological zones of Bangladesh.

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## Competing interest

The authors declare that they have no conflicts of interest that could have appeared to influence the work reported in this paper.

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