Effect of Organic and Inorganic-P Fertilizer Applications on Nitrogen Fixation and Yield of Three Mungbean Varieties in the Central of Thailand

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***Abstract*—To study the effect of organic and inorganic-P fertilizer applications on nitrogen fixation and yield of 3 mungbean varieties in the central of Thailand fields study were conducted. Three mungbean varieties as Maejo 3 (MJU 3), Khampangsan 2 variety (KPS 2) and Chainat 72 variety (CN 72) as subset experiments were designed in randomized in a Complete Block Design (RCBD) with four replications. Five treatments were laid out: 1) control (no fertilizers); 2) Triple Superphosphate (TSP) 125kg/ha (TSP Low rate); 3) TPS 250kg/ha (TSP High rate); 4) TSP Low rate with Organic Fertilizer (OF) 6.25ton/ha and 5) TSP High rate with OF 6.25ton/ha. The highest percentage of nitrogen derived from the atmosphere (%Ndfa), amount of nitrogen fixed, and yield were reported at 57.3%, 70kg N/ha and 1,138kg/ha, respectively in MJU 3 variety. TSP low rate (125kg/ha) with Organic Fertilizer (OF) 6.25ton/ha presented the high of shoot dry weight, total N, %Ndfa, amount of nitrogen fixed and yield in all varieties. In conclusion, we recommend MJU 3 variety as the best by applying TSP low rate combined with OF to increase nitrogen fixation and yield of mungbean.**

***Index Terms*—organic fertilizer, inorganic-P fertilizer, nitrogen fixation, mungbean, yield**

I. Introduction

Mungbean planted areas in central Thailand slightly declined from 138 to 128 thousand hectares in 2018 and the output was slowing down from 109 to 92 thousand tons during 2015-2016 [1]. The average yield also declined from 788 to 719 kg/ha. The domestic production was therefore far below the domestic feed and mungbean glass noodle industry demand. The total amount of import was recorded from 20 to 26 thousand tons in 2018. Consideration of farmers might be changed for many reasons such as high income with chemical fertilizer and seed production, lowing in price and lacking knowledge of mungbean production. Moreover, other countries including Myanmar, Republic of China, Australia and Pakistan could provide higher yield than Thailand. Whereas the soil fertility of mungbean production areas have many effects on their production. The imbalance of nutrient available forms is always found in the central part of Thailand. In Saraburi province, soil chemical properties are detected with high in soil pH, lack of macro and micro elements and low organic matter content [2]. Farmers get used to applying chemical fertilizers intensively for maximizing the mungbean yield and that may be the cause of unsustainable systems and lack of rhizobium inoculation.

Producing high yield and quality of seed need to have good soil quality. The optimum of nitrogen and phosphorus available is important to plant growth [3]. Hussain *et al.* [4] reported that high levels of available phosphorus in soil could increase yield when comparing with organic matter application. More confirmed this theory, Satyanarayana *et al.* [5] found that rice yield and yield quality mostly increased with the application of both organic and inorganic fertilizers in India. Generally, mungbean could fix N from atmosphere ranging from 6 to 112kg/ha [6], [7], conceding the legume plant to meet up to 75% of its N requirements from biological N_2 fixation [6]. Nonetheless, a few research in Thailand has focused on the combined effects of organic and inorganic fertilizer on mungbean plantation. The ambition of this study was to meditate the effects of inorganic-P and organic fertilizer combining with rhizobium inoculation on mungbean yield and quality. We hypothesized that plant dry matter, yield and N fixation in mungbean were varied by rate of phosphate fertilizer and organic matter content in soil property.

II. Materials and Methods

A. Experimental Design

The experiment was conducted at Prince Chakrabandh Pensiri Center for Plant Development Saraburi Province. The subset of experiment was separated by three mungbean varieties as Maejo 3 (MJU 3), Khampangsan 2 variety (KPS 2) and Chainat 72 variety (CN 72). All

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subset experiments were designed in randomized in a Complete Block Design (RCBD) with four replications and five treatments as follows: 1) control (no fertilizers), 2) Triple Superphosphate (TSP) at the low rate (125kg/ha), 3) TSP at the high rate (250kg/ha), 4) TSP at low rate plus Organic Fertilizer (OF) (6.25ton/ha) and 5) TSP at the high rate plus OF (6.25ton/ha). Seeds were inoculated with suitable native rhizobia inoculant *Bradyrhizobium elkanii* (SB2) [8]. SB2 inoculant was prepared as maximum number in excess of 10^8 CFU/g for peat-moss sterile carrier-based. Thereafter, 200g inoculants were mixed with 31kg/ha of mungbean seeds. Mungbean plant was grown in a 6×6 m-plot with 25×50 cm plant spacing. Sprinkler irrigation technique was applied during mungbean cultivation. The organic fertilizer was analyzed and calculated as: 1.83%N, 0.54%P, 0.61%K, 2.37%Ca and 1.25%Mg.

B. Sampling

Plant biomass and root bleeding sap were collected (10 plants/sample) to estimate the nitrogen fixation, nitrogen content and dry weights at the R3.5 (beginning seed) [9]. All plant samples were oven-dried at 65ºC for 48 hours and ground. The nitrogen content in the shoot was also determined by Kjedahl method. Ureide technique was used to estimate the nitrogen fixation. The relative ureide index (%RUI) was calculated from the molar concentration of ureide [10], amino [11] and nitrate [12] using the following equation.

$$\%RUI = \frac{(4 \times ureide + amino + nitrate)}{4 \times ureide} \times 100 \quad (1)$$

The percentage of nitrogen derived from the atmosphere (%Ndfa) was estimated from %RUI using the following equations [13]:

$$\%Ndfa = 1.33\,(\%RUI - 11.47) \quad (2)$$

The total N accumulation or shoot N uptake was calculated by:

$$Dry\ matter\ (g) \times (\%N)\ /100 \quad (3)$$

The amount of nitrogen fixed by mungbean was estimated from:

$$Total\ N\ accumulation \times \%Ndfa \times 1.5^* \quad (4)$$

*1.5 is a factor that is estimated for contribution by below-ground N [14].

C. Statistical Analysis

Data were analyzed for statistical significance using the Statistix 10 software analysis of variance (ANOVA), mean separation, and the Least Significant Difference (LSD) test at the significance level of P<0.05.

III. Results

According to Table I, the shoot dry weight of MJU 3 variety was indicated at R3.5 stage. It was found that application of TSP high rate + OF caused the highest shoot dry weight at 10.7 g/plant (P<0.05). The lowest shoot dry weight was found by control treatment. However, the total nitrogen was not significantly different among treatments.

Moreover, combining TSP both in high and low rate with organic fertilizer increased the %Ndfa. TSP high rate + OF had the effect of %Ndfa and nitrogen fixation higher than the other treatments at 60.2% and 83kg N/ha, respectively (P<0.05). The maximum mungbean yield was resulted by application of triple superphosphate low rate combined with an organic fertilizer at 1,194kg/ha but no significant differences with other TSP rates. However, the control treatment produced the minimum yield at 931kg/ha (P<0.05) (Table II).

TABLE I. Effect of Fertilizers Application on Shoot Dry Weight (SDW) and Total N of MJU 3 Variety

Treatment	SDW (g/plant)	Total N (%)
Control	9.0^{c}	3.43
TSP Low rate	9.2^{bc}	3.49
TSP High rate	9.7^{b}	3.54
TSP Low rate + OF	9.6^{bc}	3.52
TSP High rate + OF	10.7^{a}	3.58
Grand Mean	9.7	3.51
CV (%)	14.51	12.41
F-test	*	ns

Note: Value in the same column followed by different letters were significantly different by LSD, *=0.05.

TABLE II. Effect of Fertilizers Application on %NDFA, Amount of Nitrogen Fixed and Yield of MJU 3 Variety

Treatment	%Ndfa	Amount of nitrogen fixed (kg N/ha)	Yield (kg/ha)
Control	53.0^{c}	59^{d}	931^{b}
TSP Low rate	56.8^{b}	66^{c}	1,125^{a}
TSP High rate	57.0^{b}	71^{b}	1,213^{a}
TSP Low rate + OF	59.6^{a}	73^{b}	1,219^{a}
TSP High rate + OF	60.2^{a}	83^{a}	1,194^{a}
Grand Mean	57.3	70	1,138
CV (%)	10.58	4.96	7.82
F-test	*	*	*

Note: Value in the same column followed by different letters were significantly different by LSD, *=0.05.

Mungbean KPS 2 variety, the highest shoot dry weight was resulted by application of TSP high and low rate + OF at 9.2g/plant but not significant with only application TSP high rate (Table III). Moreover, TSP high rate + OF usually demonstrated the highest total nitrogen and amount of nitrogen fixed at 3.47% and 55kg N/ha (P< 0.05), respectively.

The percentage of nitrogen derived from the atmosphere was increased by triple superphosphate addition. The TSP high-rate treatment showed the highest of %Ndfa at 48.3 but not significantly different with TSP high rate + OF treatment (48.2%) and TSP low rate + OF (47.0%). The lowest amount of nitrogen fixation of KPS 2 was found in control treatment at 47kg N/ha but did not significantly different with TSP low rate without OF at 49kg N/ha (P<0.05). TSP low rate with organic fertilizer gave the largest grains yield at 1,100kg/ha, but not

significantly different compared to TSP low rate (Table IV). Similarly, as acted in MJU 3 variety, the control treatment detected the lowest value of yield at 775kg/ha However, an additional TPS high rate had no higher effect on the yield of KPS 2 variety (P<0.05).

TABLE III. EFFECT OF FERTILIZERS APPLICATION ON SHOOT DRY WEIGHT (SDW) AND TOTAL N OF KPS 2 VARIETY

Treatment	SDW (g/plant)	Total N (%)
Control	8.9[b]	3.17[d]
TSP Low rate	8.9[b]	3.31[c]
TSP High rate	9.1[ab]	3.35[bc]
TSP Low rate + OF	9.2[a]	3.44[ab]
TSP High rate + OF	9.2[a]	3.47[a]
Grand Mean	9.0	3.35
CV (%)	7.31	3.21
F-test	*	*

Note: Value in the same column followed by different letters were significantly different by LSD, *=0.05.

TABLE IV. EFFECT OF FERTILIZERS APPLICATION ON %NDFA, AMOUNT OF NITROGEN FIXED AND YIELD OF KPS 2 VARIETY

Treatment	%Ndfa	Amount of nitrogen fixed (kg N/ha)	Yield (kg/ha)
Control	45.7[b]	47[d]	775[c]
TSP Low rate	46.0[b]	49[cd]	1,050[ab]
TSP High rate	48.3[a]	52[bc]	981[b]
TSP Low rate + OF	47.0[ab]	53[ab]	1,100[a]
TSP High rate + OF	48.2[a]	55[a]	944[b]
Grand Mean	47.0	51	969
CV (%)	5.32	3.49	8.82
F-test	*	*	*

Note: Value in the same column followed by different letters were significantly different by LSD, *=0.05.

Dry weight of CN 72 plant biomass shown in Table V. Combining phosphorus fertilizer either low or high rate with organic fertilizer caused a significantly different shoot dry weight with other treatment that lacked organic fertilizer. The total nitrogen and %Ndfa were not significant differences among treatments. However, the highest of both parameters were presented by TSP high rate + OF at 3.46% and 38.9%, respectively. TSP high rate + OF treatment gave more amount of nitrogen fixed than other treatments at 46kg N/ha but did not differ significantly with TSP low rate + OF and only TSP high rate addition (P<0.05).

The yields of mungbean CN 72 variety were significantly different among treatments. Application TSP low rate with organic fertilizer led to a higher mungbean yield when compared with other treatments at 800kg/ha (P<0.05) but did not differ significantly with TSP high rate + OF and only TSP high rate application. However, the control treatment also detected the lowest yield at 450kg/ha (Table VI).

Referring to the best variety of mungbean that caused the highest yield in this study, the cost benefit analysis was examined only in MJU 3 variety. The explanation for the usage of inorganic and organic fertilizers is due to the cost benefit analysis (Table VII), which is provided the optimum as a low rate of TSP (125kg/ha) combined with organic fertilizer (6.25ton/ha). It can calculate that the highest yield with a good profit (7,585 Thai baht/ha) was earned by TSP low rate + OF. Application TSP high rate (250kg/ha) gave the highest profit cost (8,190 Thai baht/ha) with 4,500 Thai baht of total fertilizer cost, but it may cause the long term side effect of soil fertility in mungbean production areas. At present, the TSP fertilizer is not available in Thailand then to increase phosphorus content in soil instead by organic P (compost, manure, biofertilizer, etc.) or inorganic fertilizer as other formulas.

TABLE V. EFFECT OF FERTILIZERS APPLICATION ON SHOOT DRY WEIGHT (SDW) AND TOTAL N OF CN 72 VARIETY

Treatment	SDW (g/plant)	Total N (%)
Control	8.9[c]	3.12
TSP Low rate	9.0[bc]	3.25
TSP High rate	9.1[bc]	3.44
TSP Low rate + OF	9.3[ab]	3.37
TSP High rate + OF	9.6[a]	3.46
Grand Mean	9.2	3.33
CV (%)	8.52	5.31
F-test	*	ns

Note: Value in the same column followed by different letters were significantly different by LSD, *=0.05.

TABLE VI. EFFECT OF FERTILIZERS APPLICATION ON %NDFA, AMOUNT OF NITROGEN FIXED AND YIELD OF CN 72 VARIETY

Treatment	%Ndfa	Amount of nitrogen fixed (kg N/ha)	Yield (kg/ha)
Control	33.9	34c	450c
TSP Low rate	34.4	36bc	619b
TSP High rate	36.5	41ab	775ab
TSP Low rate + OF	36.4	41ab	800a
TSP High rate + OF	38.9	46a	706ab
Grand Mean	36.0	40	669
CV (%)	9.26	11.34	11.06
F-test	ns	*	*

Note: Value in the same column followed by different letters were significantly di fferent by LSD, *=0.05.

TABLE VII. COST BENEFIT ANALYSIS OF MAEJO 3 VARIETY PRODUCTION

Variable	CF cost	OF cost	Total fertilizer cost	Benefit cost	Net return	Profit
	(----------Thai Baht/ha----------)					
Control	-	-	-	41,895	41,895	-
TSP Low rate	2,250	-	2,250	50,625	48,375	6,480
TSP High rate	4,500	-	4,500	54,585	50,085	8,190
TSP Low rate + OF	2,250	3,125	5,375	54,855	49,480	7,585
TSP High rate + OF	4,500	3,125	7,625	53,730	46,105	4,210

CF = Chemical fertilizer, OF = Organic fertilizer
*Benefit cost = Yield × Present price, Net return = Benefit Cost-Total fertilizer cost, Profit = Price increasing from control

IV. Discussion

Shoot dry weight showed an increase by the TSP high rate + OF but not significantly different compared to TSP low rate + OF all varieties. These results are similar to the findings of Suryantini [15] who studied mungbean grown after soybean planting. The results revealed that plant dry weight and grain yield were increased by the application of organic and high levels of phosphorus fertilizer. Total N in shoot were not significant differences in MJU 3 and CN 72 varieties but differed significantly in KPS 2. In our study KPS 2 variety gave the higher percentage of nitrogen than the reported of Boonked *et al.* [16] who found the percentage of nitrogen at 2.40%N at maturity stage. Moreover, applying TSP high-rate combination with or without organic fertilizer could enhance nitrogen uptake in shoot. Similar to the report of Hussain *et al.* [17], [18] they found that the maximum nitrogen and phosphorus uptake in plants varied with the highest dose of P at 150% recommended. The percentage of nitrogen derived from the atmosphere (%Ndfa) and amount of nitrogen fixed in the mungbean were strongly influenced by mungbean genotypes. In our study MJU 3 gave the best of %Ndfa and amount of nitrogen fixed compared with KPS 2 and CN 72. However, KPS 2 recorded the %Ndfa average at 47.0% this agrees with the report of Boonked *et al.* [19]. They reported that the %Ndfa of KPS 2 ranged from 49.57 to 54.05% (64 days after planting). Furthermore, the average amount of nitrogen fixed of CN 72 in this experiment was determined at 39.7 kg N/ha (R3.5 stage). Previous study showed the amount of nitrogen fixed at 23.9kg N/ha (post-harvest) [20]. Phoomthaisong *et al.* [21] reported that %Ndfa by ^{15}N isotope dilution method and amount of nitrogen fixed in six mungbean cultivars ranged from 54% to 62% and 35-50kg N/ha. In this experiment, the %Ndfa and amount of nitrogen fixed were enhanced by either only TSP or TSP combined with organic fertilizer application. The result liked the view that %Ndfa and amount of nitrogen fixed depended significantly on phosphorus requirements of three mungbean cultivars [22].

Mungbean yields, MJU 3, KPS 2 and CN 72 showed the average at 1,219, 1,100 and 800kg/ha. The result related with Ngampongsai *et al.* [23] who reported the KPS 2 yield at 900-1,106 kg/ha for dry and rainy reasons, respectively. The overall yield of this experiment was higher than the result of Verma *et al.* [24] with 597.3kg/ha. Moreover, Phoomthaisong [25] recommended applying 37.5kg P_2O_5/ha with rhizobium inoculation on mungbean KPS 2 variety could provide the higher N fixation and yield at east north of Thailand. The combining organic and phosphate fertilizer increased the number of seeds per pod [26]. Not only mungbean but cowpea could respond with triple superphosphate on the amount of seed per pod, 100-seed weight and yield higher than single superphosphate and nitrophoska (15-15-15). Interestingly, the high rate of phosphate fertilizer provided a high amount of pod per plant, seed per pod, 100-seed weight, and yield [7], [27].

V. Conclusion

In summary, the nitrogen fixation capacity of the mungbean was enhanced by application of both phosphorus and organic fertilizer at the central part of Thailand. The result clearly demonstrated that TSP low rate plus organic fertilizer provided the highest yield in all mungbean varieties, especially in MJU 3. More comprehensive and detailed studies should be carried out to amend the yield and quality of mungbean under low phosphorus available and the percentage of organic matter in the field condition of the middle region, Thailand.

Conflict of Interest

The authors declare no conflict of interest.

Author Contributions

C. Dechjiraratthanasiri and J. Inthasan carried out the experiment. C. Dechjiraratthanasiri wrote the paper with support from J. Inthasan and C. Santasup. All authors had approved the final version.

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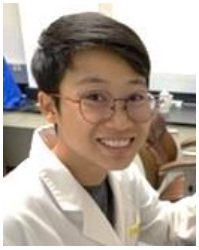

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