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Effect of Using Agro-fertilizers and N-fixing Azotobacter Enhanced Biofertilizers on the Growth and Yield of Corn

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Abstract: Corn is an important crop and is grown widely around the world. Corn is a food source for human as well as animal and processed into industrial product such as ethanol. Corn is one of the important productions of Malaysia as the climate is suitable for corn growth. In this study, organic fertilizers such as biofertilizer, N-fixing Azotobacter enhanced biofertilizers and compost were added to the soil to determine best practice in using organic fertilizers for higher corn yield and growth. The study was conducted in plot experiment with five replications based on randomized block design in the summer of 2012. All plots were manually harvested and yield was adjusted to 15% moisture. Grain yield (total corn harvested) at maturity was determined by harvesting the two central rows of each plot. Statistical analysis was performed on the effect of fertilizer treatments on plant growth, corn yield and nitrogen, phosphorous and potassium contents of plant materials. The means were compared according to Duncan multiple range test. The results showed that organic fertilizers in the form of N-fixing Azotobacter enhanced biofertilizer increased yield with positive effects on measured plant height, weight and leaf index. Given the significant enhancement in growth and yield of corn taking place mainly with N-fixing Azotobacter fertilizers under organic condition, the mechanism for this beneficial effect could be due to the more balanced nutrition and improved absorption of nitrogen and other mineral nutrients by the corn.

Key words: Biofertilizer, N-fixing Azotobacter, compost, fertilizer treatments, corn yield

INTRODUCTION

Agricultural crops require numerous essential elements for satisfactory crop growth and production. Carbon, hydrogen and oxygen are non-mineral elements derived from the atmosphere and water. The mineral elements which arise from the soil are dissolved in water and absorbed through a plant's roots to complete the crop's life cycle. Plants need all these mineral elements to support their life cycle by absorbing different amounts of these elements into plant tissues. Macronutrients which include nitrogen, phosphorus, potassium, calcium, magnesium and sulphur are required in large amount which occupy 0.2 to 0.4% of the dry weight mass of plant tissues. Micronutrients (iron, copper, manganese, zinc, boron, molybdenum and chlorine) are required in relatively smaller amounts, ranging from 5 to 200 ppm, or less than 0.02% dry weight (Mills and Jones, 1996). However, there are not always sufficient nutrients present in the soil for a plant to consume for healthy and productive growth. To compensate the deficiency of nutrients in the soil, fertilizers are added to the soil during plant growth. There are two types of fertilizer: organic fertilizer and inorganic or chemical fertilizer.

Chemical fertilizers are a mixture of various chemicals made to enhance the three major types of primary macronutrients namely: nitrogen, phosphorus and potassium concentration in the soil. In Malaysia, local farmer rely heavily on chemical fertilizer (90%) out of all fertilizers used throughout the farming systems. In an effort to reduce the dependence on chemical fertilizers and to move towards more natural and healthier approach of food production, the government is promoting programs that encourage the recycling and composting of agricultural waste for organic fertilizer (MOA, 2012). Organic fertilizers are naturally occurring fertilizers derived from composting process of organic materials. Organic fertilizer contains nitrogen, phosphorus, potassium and other trace element needed to help plants grow successfully (Nino *et al*., 2012; Chand *et al*., 2006). Although, the density of nutrients in organic material is comparatively modest, nonetheless, the majority of organic fertilizer contains insoluble nitrogen and can act as a slow-release fertilizer (Bokhtiar and Sakurai, 2005).

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Organic fertilizers help improve the physical and biological nutrient storage mechanisms in soil naturally and thus assist in coping the risk of over-fertilization. In this study, organic fertilizers was the main focus for corn production and were added to the soil in the form of biofertilizer, N-fixing enhanced biofertilizer and compost to determine the best application of organic fertilizers for higher corn yield and growth. Corn is one of the important food crops in this country. Corns require adequate nutrients for their growth. It needs large amount of nitrogen (N), phosphate (P_2O_5), potash (K_2O) whereas secondary nutrient (calcium, magnesium and sulphur) and micronutrients are needed in small quantity. With that said, proper soil fertility is a key to profitable corn production (Shanti *et al*., 1997; Vos *et al*., 1998). Secondary macronutrients are available in the market but not many corn farmers in Malaysia use these fertilizers (FAO, 2004). Although micronutrients are needed in small quantities, but insufficient of it will affect the growth of corn (Barbosa *et al*., 2012). These micronutrients can be supplied through the organic fertilizers to the soil. Corn yields often face the lack of nitrogen supply instead of deficiencies of other essential nutrients (Mikkelsen and Hartz, 2008).This is because corn has a high nitrogen requirement and depletion of nitrogen supplied can occur during the growing season by leaching, denitrification or other processes. Nitrogen promotes protein and chlorophyll, hence adequate amount of it can stimulate the growth of stems and leaves. Effects of the addition of N-fixing bacteria Azotobacter as N enhancer for corn growth and production was also evaluated in this study. Azotobacter was reported by many researchers regarding its ability to transform the chemically stable gas nitrogen from atmosphere into organic nitrogen compounds and consequently enhance the nitrogen content in the soil (Aquilantia *et al*., 2004; Bashyal, 2011).

Proper fertilization is required to achieve optimum corn production. The profit potential for farmers depends on producing enough corn per acre to keep the production costs below the selling price. One of the important key to profitable corn yield is meeting the nutrient supply level by applying the right types and amounts of fertilizers efficiently. In this study, field studies were conducted to investigate the use N-fixing Azotobacter enhanced biofertilizers and compost on the NPK contents of plant materials. The effect of fertilizers on the plant biomass, growth and yield of corn was also investigated.

MATERIALS AND METHODS

The corn trial was carried out during the period of 13 June 2012-30 August 2012 (11 weeks).

Materials: The fertilizers used in this study were supplied by All Cosmos Industries Sdn Bhd. The compost was prepared in Faculty of Forestry, Universiti Putra Malaysia. Matching fertilizer application rates to corn needs is an essential component of optimizing corn production. However, different crops in separate fields will require varying rates of the major nutrients-N, P_2O_5 and K_2O. In this study, the fertilizer rates applied for the corn as shown in Table 1 were based on the soil types for the corn trial and total N required by corn. The soil type is typic paleudult and is known as the Rengam Series, an inland sedentary soil from granitic parent materials.

Methods: The experimental design was a randomized complete block. The treatment consisted of 5 different sources of fertilizer applied. The fertilizer sources are chemical, organic, N-fixing enhanced biofertilizers and the control (no application).

The timing of fertilizer applications is scheduled to apply fertilizer as close as possible to the period of rapid crop uptake. Managing fertilizer in this way will minimize losses of nitrogen from the field and will ensure adequate nitrogen availability for the corn during critical growth periods. The fertilizers were applied in 5 split doses. The starter dose was applied basally at planting stage and the second application of fertilizers was top dressed about a week after planting (emergence stage). Third dose of fertilizers were broadcasted after 4 weeks of planting. Forth dose of fertilizers were applied when silks have emerged from the tip of the ear shoot on at least 50% of the plants. Last dose of fertilizers were broadcasted after 14 days of silking. Due to the slow release nature of organic fertilizer, compost was applied a week before planting while chemical and biochemical fertilizers were applied at planting.

The experimental field was divided into plots. Each plot measured 2×5 m with spacing of 0.5 m between each plot and fitted with five replications based on randomized block design. The planting spacing used for the experiment was 75 cm between rows and 25 cm within rows. Two seeds were planted per hole and later thinned to one plant per stand. Weeds were controlled with a combination of pre-emergence and contact herbicides applied at planting, followed by hand weeding at 3 weeks and 7 weeks after planting.

All plots were manually harvested and yield was adjusted to 15% moisture. Grain yield (total corn harvested) at maturity was determined by harvesting the two central rows of each plot. Random samples consisting of 10 ears per plot were taken for the determinations of weight of corn at harvest. In addition total dry matter of plant maturity was measured. Plant height (extended leaf)

Table 1: Type of fertilizer and application rate of treatment

Treatment	Type of fertilizer	Application stage	NPK rating	Application rate (kg/acre)
1	Control (without fertilizer)	-	-	-
2	Commercial Inorganic NPK fertilizer	Starter	15:15:15	150
		Second application	15:15:15	150
		Third application	15:15:15	150
		Forth application	12:12:17	200
		Fifth application	12:12:17	150
3	Biofertilizer (Minimum 30% composted organic	Starter	5:05:05	500
	materials mixed with chemical fertilizers)	Second application	11:11:11	150
		Third application	11:11:11	150
		Forth application	8:08:20	200
		Fifth application	8:08:20	150
4	N-fixing Azotobacter Enhanced Biofertilizers	Starter	5:05:05	500
	(106 CFU per gram of the biofertilizer)	Second application	11:11:11	150
		Third application	11:11:11	150
		Forth application	8:08:20	200
		Fifth application	8:08:20	150
5	Urea	Starter	46:00:00	25
		Second application	46:00:00	16.5
		Third application	46:00:00	16.5
		Forth application	46:00:00	16
		Fifth application	46:00:00	12
6	N-fixing Azotobacter Mixed with Compost made from	Starter	1.4:1.1:2.8	500
	Coffee ground and cocoa shell	Second application	1.4:1.1:2.8	330
	(106 CFU per gram of the biofertilizer)	Third application	1.4:1.1:2.8	330
		Forth application	1.4:1.1:2.8	320
		Fifth application	1.4:1.1:2.8	240

was recorded after harvesting. Area of green leaf was measured using a leaf area meter and aboveground plant parts were separated into stems and leaves. All data were converted to a common format (yield/hectare, biomass/ha and total corn/ha). Harvest Index of ratio of the economical yield (grain) to total aboveground biomass was also presented in this study.

The harvested plant materials (root, stem and leaves) were wet digested by weighing 0.2 g of each material into a separate 50 mL digesting tube. Total N was analyzed on elemental CNS analyzer Vario EL III (Nelson and Sommers, 1996). Total phosphorous and potassium contents of the plant materials were determined according to the method described by Karla (1998).

Statistical analysis: Statistical analyses were performed using SAS computer program. The means were compared according to Duncan multiple range test. An effect was considered to be significant if its p-value was = 0.05. All these experiments were performed in five replications.

RESULTS AND DISCUSSION

The results as indicated in Table 2 showed that Azotobacter enhanced biofertilizer and compost application increased plant biomass, corn weight, grain yield and harvest index compared to those straight and compound fertilizers. Table 3 shows the Azotobacter enhanced biofertilizer and compost significantly increased NPK content of plant materials. Analysis of variance showed that there were significant differences among all treatments on plant growth, corn yield and NPK content of plant materials.

Plant growth and corn yield: The main effects of agro-fertilizers and N-fixing Azotobacter Enhanced Biofertilizers on plant and corn are presented in Table 2. Results showed that Treatment 4 (N-fixing Azotobacter Enhanced Biofertilizer) had significant effect on plant growth and yield of corn.

The maximum leaf area (6314 cm^2) and weight of plant (113.8 g) were recorded at treatment 4. With such results, the increasing of leaf area enhances photosynthesis rate and this enhancement leads to increasing yield. Shanthi *et al.* (2012) had reported that N fixing Azotobacter is able to supply high amount of nitrogen for tissue growing and therefore increases chlorophyll content. Due to the main effects of nitrogen, treatment 4 and their interactions on corn parameter were highly significant (Table 2) with corn biomass (21767 kg ha^{-1}), total corn (271 unit ha^{-1}), yield (8775 kg ha^{-1}) and harvest index (0.403). Harvest index is a measure of success in partitioning photosynthetic to harvestable product. This result confirmed that compost with Azotobacter at suitable application rate can improve significantly the growth of corn. Kader *et al.* (2002) reported that N-fixation bacteria that live in Rhizosphere enhance root growth by stimulating auxin and gibberellins hormone that cause

Table 2: Plant biomass and corn yield components

	Plant						Corn		
Treatment	Height (cm)	Total Leaf area (cm^2)	Weigh leaf ($cm^2\ g^{-1}$)	[1]SLA plant ($cm^2\ g^{-1}$)	[2]LAR ($kg\ ha^{-1}$)	biomass	Total corn Unit/ha	Grain yield ($kg\ ha^{-1}$)	Harvest Index
1	159^{c}	2004^{f}	45.0^{e}	175^{d}	44.5^{b}	8474^{e}	79^{e}	1824^{e}	0.215^{d}
2	191^{a}	5017^{d}	90.8^{d}	269^{c}	55.3^{a}	13841^{d}	175^{c}	4500^{d}	0.325^{c}
3	184^{ab}	5453^{b}	99.1^{b}	282^{ab}	55.0^{a}	17904^{b}	217^{b}	6464^{b}	0.362^{b}
4	184^{ab}	6314^{a}	113.8^{a}	291^{a}	55.5^{a}	21767^{a}	271^{a}	8775^{a}	0.403^{a}
5	189^{ab}	4907^{e}	90.1^{d}	270^{c}	54.5^{a}	13269^{d}	155^{d}	4310^{d}	0.325^{c}
6	183^{b}	5255^{c}	95.7^{c}	279^{bc}	54.9^{a}	16272^{c}	209^{b}	5789^{c}	0.570^{b}

Note: Means with the same letter on each column are not significantly different, [1]SLA refers to specific leaf area; [2]LAR refers to leaf area ratio

Table 3: NPK contents of corn leaves, stems and roots at 11 weeks after planting

	Nitrogen (N) %			Phosphorous (P2O5) %			Potassium (K2O)%		
Treatment	Leaves	Stems	Roots	Leaves	Stems	Roots	Leaves	Stems	Roots
1	2.35^{e}	1.63^{e}	1.92^{e}	0.16^{c}	0.09^{d}	0.09^{c}	1.00^{d}	0.91^{d}	1.18^{f}
2	3.02^{b}	2.76^{b}	2.60^{c}	0.21^{b}	0.23^{a}	0.18^{a}	2.06^{b}	1.35^{b}	1.78^{c}
3	3.13^{b}	2.88^{a}	2.92^{b}	0.26^{a}	0.23^{a}	0.18^{a}	2.37^{a}	1.43^{a}	2.28^{b}
4	3.55^{a}	2.92^{a}	3.61^{a}	0.26^{a}	0.24^{a}	0.18^{a}	2.39^{a}	1.45^{a}	2.41^{a}
5	2.67^{d}	2.48^{d}	2.25^{d}	0.20^{b}	0.12^{c}	0.12^{b}	1.06^{d}	0.93^{d}	1.50^{e}
6	2.80^{c}	2.62^{c}	2.21^{d}	0.20^{b}	0.15^{b}	0.12^{b}	1.33^{c}	1.08^{c}	1.64^{d}

Note: Means with the same letter on each column are not significantly

an increase in nutrient uptake from soil that influence plant vegetative and reproductive growth. Biari *et al.* (2008) also found that bacteria like Azospirillum and Azotobacter as growth stimulators have optimistic effects on corn yield when grown at field under organic conditions. Treatment 1 (control, without fertilizer), was recorded with the lowest height, leaf area, weight, SLA, LAR, biomass, total corn yield and harvest index as follow, 159 cm, 2004 cm^2, 45.0 g, 175 $cm^2\ g^{-1}$ leaf, 44.5 $cm^2\ g^{-1}$ plant, 8474 $kg\ ha^{-1}$, 79 unit ha^{-1} 1824 $kg\ ha^{-1}$ and 0.215, respectively.

NPK contents of Plant materials: Table 3 shows the percentage of NPK in plant parts. Treatment 4 recorded the highest N percentage in the leaves and the lowest N was found in the control treatment. For the stems and roots, N percentage was the highest in T4. The percentage of N was higher in the leaves compared to that in the roots and stems. A total N (%) concentration of 2.5 to 2.7 in the ear leaves indicates that N supply is adequate for optimum corn yield (Fox *et al.*, 2001). The result has shown significant difference ($p = 0.05$) among the treatments in respect to roots, stem and leaves for phosphorous (P_2O_5) percentage. In this study, it was shown that T3 and T4 yielded the highest concentration of Phosphorous in the leaves. For the stem, the highest concentration was observed in T2 (0.23%). Treatment 2, T3 and T4 recorded the highest phosphorous in the roots (0.18%) while the lowest phosphorous was found in T1. In the case of potassium concentration in plant parts, the highest percentage in the leaves, stems and roots was discovered in T4. Treatment 1, control showed the lowest concentration for all plant parts. The least values of growth parameters of corn in the control treatment could be traced to the initial poor nutrient status of the soil. NPK are the essential nutrients required for the meristematic and physiological activities of leaves, roots, shoots and dry matter production. According to Table 3, plants contain more NPK with fertilizers treatment compared to the control treatment. This indicated that, application of NPK fertilizer could improve the nutrient uptake of plants (Law-Ogbomo and Law-Ogbomo, 2009). However, the effect of treatment 4 was significantly different from other treatments due to the addition of N-fixing Azotobacter. Azotobacter plays an important role in nitrogen cycle, binding nitrogen which is inaccessible to plants and releasing it in the form of ammonium ions into the soil (Gandora *et al.*, 1998). Furthermore, composted organic materials showed better plant growth than inorganic NPK fertilizer. It may be due to the beneficial effects of compost in supplying plant nutrients, improving the cation exchange capacity that enables them to retain nutrients longer. Improving the cation exchange capacity especially in sandy soils can greatly improve the retention of plant nutrients in the root zone and also supporting soil biological activities (Sarker *et al.*, 2012).

CONCLUSION

The organic fertilizers in the form of biofertilizer and compost enhanced with N-fixing Azotobacter apparently improved nutrient uptake and could have explained the improvement of plant growth and yields of corn in this study. The mechanism of this beneficial effect could be due to the fact that composts are suitable carriers for Azotobacter. Such combination provides the plant with more balanced nutrition. It also improves the absorption of NPK as well as other mineral nutrients from the compost that are essential for corn plants to grow and achieve optimum yield.

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