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Full Length Research Paper

Effect of N, P and K humates on dry matter of *Zea mays* and soil pH, exchangeable ammonium and available nitrate

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Ammonia volatilization from surface-applied urea reduces urea-N use efficiency in crop production and it also pollutes the environment; it is an economic loss. A greenhouse study was conducted to confirm the effect of similar fertilizer formulations (N, P and K humates) on soil pH, exchangeable ammonium, available nitrate retention and dry matter of *Zea mays* cultivated on an acid soil (Typic Paleudults). The fertilizers were applied 10 days after planting (DAP) in each pot containing 10 kg of soil. Soil and plant samples (stems, leaves and roots) were collected at 31 DAP. Soil samples were analyzed for pH, ammonium, and nitrate content. Urea amended with humic acid (HA), acidified (HA+FA) and humin without TSP and MOP were not effective in increasing the dry matter production of the test crop. Urea amended with fulvic acid (FA) alone significantly increased plant dry matter. Complete fertilizer consisting urea, triple superphosphate (TSP) and mono triphosphate (MOP) amended with or without HA, FA, acidified HA and FA and humin significantly increased the dry matter of the test crop with significant retention of soil exchangeable ammonium. However, only the complete fertilizer with and without HA and humin amendment significantly retained soil available nitrate. The findings in this study may only be applicable to similar acid soils. The outcome of this study may contribute to the improvement of urea N use efficiency as well as reducing environmental pollution.

Key words: Humic acids, fulvic acids, triple superphosphate, muriate of potash, soil exchangeable ammonium, available nitrate, *Zea mays*, dry matter.

INTRODUCTION

Volatilization from surface applied urea causes a significant loss of N (Prasertsak et al., 2001; Cai et al., 2002) and ammonia into the atmosphere. Ammonia volatilization is serious when it occurs in acid soils having low pH, cation exchange capacity and basic cations contents (Syed, 2001). Ninety three percent of tropical soils are known to have nitrogen (N) deficient problem. Nitrogen fertilizer is of particular importance because it rapidly undergoes denitrification, volatilization or leaching while phosphates and potash are largely fixed or accumulated in the soils if they are not removed by the plants (Soh, 2001).

The prevalent concern over N fertilizer losses has led to various researches that have revealed the role of humic acids (HA), fulvic acids (FA) and triple superphosphate (TSP) as amendments to delay hydrolysis of urea fertilizer, thereby controlling ammonia volatilization. Low pH and high total acidity (cation exchange capacity, CEC) associated with HA and FA enable them to inhibit

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Abbreviations: DAP, Days after planting; **TSP,** triple superphosphate; **MOP,** mono triphosphate; **HA,** humic acid; **FA,** fulvic acid.

urease activity, retain ammonium, delay hydrolysis of urea, thereby reducing ammonia volatilization (Latifah et al., 2010, 2011; Rosliza et al., 2009a, b; Ameera et al., 2009; Regis et al., 2009; Susilawati et al., 2009a; Ahmed et al., 2003, 2008a, 2006a, b). According to Bock and Kissel (1998), the ability of ureaphosphate (mixtures of urea with TSP) to inactivate soil urease causes ammonia volatilization reduction because of H_3PO_4 from the mixture. Mixing soluble salts of K (KCl) with urea fertilizer may also reduce ammonia loss.

In laboratory studies, varying degrees of significant reduction of ammonia volatilization has been reported when urea was amended with humic substances, TSP, and MOP (Rosliza et al., 2009a, b). However, these studies were carried out under laboratory conditions; as such, the results may not reflect real effects on plants. Therefore, this study was conducted to evaluate the effects of urea amended with HA, FA, TSP or MOP on soil pH, exchangeable ammonium, available nitrate and total dry matter production (stems, leaves and roots) of an improved Masmadu maize variety (*Zea mays*) cultivated on an acid soil (Typic Paleudults) under greenhouse conditions.

MATERIALS AND METHODS

The soil used in this study was a sandy loam, classified as Typic Paleudults (Bekenu Series) collected from Universiti Putra Malaysia, Bintulu Sarawak Campus, Malaysia, at a depth of 0 to 15 cm. The palm oil mill effluent (POME) sludge samples used in this study were collected randomly from Usaha Sepadan Palm Oil Mill, Bintulu Sarawak, Malaysia. Both soil and POME sludge were air dried after which they were ground and sieved to pass a 2 mm sieve for further analysis and the glasshouse experiment. The selected chemical and physical properties of the soil were determined using standard procedures. The soil pH was determined in a 1:2.5 (soil: distilled water) suspension and 1 M KCl using a digital pH meter. Soil organic carbon was determined as 58% of the total loss of weight on ignition (Cheftez et al., 1996). The hydrometer method was used to determine soil texture (Tan, 2005). The leaching method was used to determine cation exchange capacity (CEC) (Cottenie, 1980). The exchangeable cations (K, Ca, Na and Mg) were extracted using the double acid method after which they were determined using atomic absorption spectrophotometry. Total N was determined using Kjedhal method (Tan, 2005).

HA and FA were isolated from air-dried POME sludge samples using the method described by Stevenson (1994) with some modifications (Ahmed et al., 2004). The extraction and fractionation periods used were 24 h. HA and FA was isolated using 0.5 M KOH and 6 N H_2SO_4, respectively. Total organic carbon of HA was determined using the loss on ignition method (Cheftez et al., 1996). The carboxylic-COOH, phenolic-OH functional groups and total acidity of HA were determined using the method described by Inbar et al. (1990). The E_4/E_6 of HA was determined using E_4/E_6 ratio (Tan, 2005). The solid HA was ground and sieved through 250 µm aperture and the urea was in granular form. FA and (HA+FA) were in liquid form.

A greenhouse study was conducted using a randomized complete block design (RCBD) with 3 replications. Treatments evaluated are as follows: soil only (control) (T0), 7.32 g urea (control) (T1), 7.32 g urea + 2.72 g HA (T2), 7.32 g urea + 7.32 g TSP + 0.6 g MOP + 2.72 g HA (T3), 7.32 g urea + 7.32 g TSP + 0.6 g MOP (T4), 7.32 g urea + 217 mL FA (T5), 7.32 g urea + 7.32 g TSP + 0.6 g MOP + 217 mL FA (T6), 7.32 g urea + 217 mL acidified (HA+FA) (T7), 7.32 g urea + 7.32 g TSP + 0.6 g MOP + 217 mL acidified (HA+FA) (T8), 7.32 g urea + 300 g humin (T9), 7.32 g urea + 7.32 g TSP + 0.6 g MOP + 300 g humin (T10).

Thirty plastic pots were filled with 10 kg of soil each. The fertilizer requirement of improved Masmadu (*Zea mays*) was scaled down to 3 plants per pot (7.32 g pot^{-1} urea; 7.32 g pot^{-1} TSP; 0.60 g pot^{-1} MOP) based on the requirement of 53,333 plants per hectare (130 kg ha^{-1} urea; 130 kg ha^{-1} TSP; 100 kg ha^{-1} MOP). The fertilizers were surface applied ten days after planting (10 DAP).

The plants were harvested at the tassel stage, because this stage is the maximum growth stage the plant can achieve before commencing the productive stage. The shoots of the plants were harvested and partitioned into leaf and stem. The roots were collected by washing the soil from the roots using tap water. These plant parts were oven dried at 60°C to constant weight and after which they were weighed using a digital balance.

A day before harvesting, soil samples were taken from the pots and analyzed for pH, exchangeable NH_4^+ and available NO_3 using standard methods. Analysis of variance (ANOVA) was used to test treatment effects while treatments means were compared using Tukey's Test (SAS, 2001).

RESULTS AND DISCUSSION

The selected soil chemical properties (Table 1) were typical of the Bekenu Series and were consistent with those reported by Paramanathan (2000), except for the high values of pH, organic carbon, CEC and exchangeable calcium. The pH of urea was high as expected while that of HA and FA was low. The carbon, carboxylic, phenolic, total acidity and E_4/E_6 values of the HA were within the range reported by Stevenson (1994) and Tan (2003).

Treatments T3, T4, T5, T6 and T8 (excluding T10) increased dry matter production (DMP) of the test crop and also showed significant reduction of ammonia volatilization reported in a previous laboratory study (Tables 2 and 3).

The DMP of the test crop fertilized with urea mixed with TSP, MOP and solid HA (T3) was higher than the control (T1) as shown in Table 3. T3 also significantly increased soil pH, exchangeable ammonium and available nitrate retention compared to the control (Table 4).

The second best DMP of the test crop was for the mixture of urea TSP, MOP and liquid FA (T6) with the highest retention of soil exchangeable ammonium and lower value of soil pH compared to the control (Tables 3 and 4). However, soil available nitrate was not significantly different from the control (Table 4).

The third best DMP of the test crop was for the mixture of urea, TSP, MOP and acidified (HA+FA) (T8) with the second best retention of soil exchangeable ammonium and higher value of soil pH compared to the control

Table 1. Some chemical and physical characteristics of soil, urea, humic acid (HA), fulvic acid (FA) and acidified (HA + FA).

Property	Soil	Urea	HA	FA	Acidified (HA+FA)
pH (water)	6.32	8.06	nd	1.13	1.00
pH (N KCl)	5.52	nd	nd	nd	nd
Total organic carbon (%)	4.72	nd	54.95	nd	nd
Nitrogen (%)	0.17	nd	nd	nd	nd
CEC (cmol kg^{-1})	13.3	nd	nd	nd	nd
Exchangeable K (cmol kg^{-1})	0.18	nd	nd	nd	nd
Exchangeable Ca (cmol kg^{-1})	1.21	nd	nd	0.89	nd
Exchangeable Na (cmol kg^{-1})	0.01	nd	nd	2.78	nd
Exchangeable Mg (cmol kg^{-1})	0.12	nd	nd	0.29	nd
Texture	LS	nd	nd	nd	nd
Carboxylic group (cmol kg^{-1})	nd	nd	538.81	nd	nd
Phenolic group (cmol kg^{-1})	nd	nd	293.89	nd	nd
Total acidity(cmol kg^{-1})	nd	nd	832.70	nd	nd
E_4/E_6	nd	nd	8.02	nd	nd

CEC: Cation exchange capacity; LS: Loamy sand; nd, not determined; [a]: CEC of HA = total acidity.

Table 2. Total amount and reduction of NH_3 percentage loss from a previous laboratory study (Rosliza et al., 2009a, b).

Treatment	Total amount of NH_3 loss (%)	Reduction of NH_3 loss (%)
(T0) Soil	0.00^{e}	-
(T1) Urea only	48.21^{a}	-
(T2) Urea + HA	46.32^{a}	$4\%^{(ns)}$
(T3) Urea + TSP + MOP + HA	41.98^{b}	13
(T4) Urea + TSP + MOP	38.51^{c}	20
(T5) Urea + FA	37.05^{c}	23
(T6) Urea + TSP + MOP + FA	33.97^{d}	30
(T7) Urea + HFA	0.00^{e}	100
(T8) Urea + TSP + MOP + HFA	0.00^{e}	100
(T9) Urea + humin	-	-
(t10) urea + tsp + mop + humin	-	-

Different letters within a column indicate significant difference between means using Tukey's test at p = 0.05; ns, non significant

(Tables 3 and 4). However, soil available nitrate was not significantly different from the control (Table 4).

The fourth best DMP of the test crop was for the mixture of urea, TSP, MOP and humin (T10) with significantly higher retention of soil exchangeable ammonium, available nitrate and highest value of soil pH compared to the control (Tables 3 and 4).

The fifth best DMP of the test crop was for the mixture of urea and FA (T5) with significant retention of soil exchangeable ammonium, available nitrate and the lowest value of soil pH compared to the control (Tables 3 and 4).

The sixth best DMP of the test crop was for the mixture of urea, TSP and MOP (T4) wherein the real effect of HA, FA and humin is apparent (Table 3). Soil available nitrate was low only for the mixtures of urea, TSP, MOP and acidified (HA+FA) (T8) (Table 4). Soil pH was lower for the mixtures of urea, TSP and MOP (T4), urea amended with FA (T5) and urea amended with FA, TSP and MOP (T6) while the other formulations of urea amended with TSP, MOP and HA (T3), urea amended with TSP, MOP and acidified (HA+FA) (T8) and urea amended with TSP, MOP and humin (T10) were higher than that of the control (Table 4).

The seventh best DMP of the test crop was for urea alone (T1). From the foregoing results, 6 treatments improved total DMP of the test crop compared to the control (T1) in the order starting T4 < T5 < T10 < T8 < T6 < T3. Mixtures that failed to improve DMP of the test crop after urea were in the order T2 > T7 > T0 > T9 (Table 3). For the mixtures of urea amended with acidified (HA+FA), the DMP of the test crop was not as high as expected though it doubled soil exchangeable ammonium and caused significantly higher available nitrate compared to the control (Tables 3 and 4). Soil pH was found to be significantly lower than the control (Table 4).

Table 3. Total plant dry weight (stems, leaves, roots) of *Zea mays* at 31 days after planting (DAP).

Treatment	Total plant dry weight (g)
(T0) Soil	3.27[j]
(T1) Urea only	5.89[g]
(T2) Urea + HA	5.53[h]
(T3) Urea + TSP + MOP + HA	12.41[a]
(T4) Urea + TSP + MOP	6.50[f]
(T5) Urea + FA	8.04[e]
(T6) Urea + TSP + MOP + FA	11.28[b]
(T7) Urea + HFA	4.76[i]
(T8) Urea + TSP + MOP + HFA	10.05[c]

Different letters within a column indicate significant difference between means using Tukey's test at p = 0.05.

Table 4. Effect of N, P, and K humates on soil pH, available nitrate and exchangeable ammonium under laboratory (Rosliza et al., 2009a, b) and greenhouse conditions.

Treatment	pH (H_2O)	NO_3 (ppm)	NH_4 (ppm)
Previous Laboratory study			
(T0) Soil	6.7[d]	11.68[c]	81.73[e]
(T1) Urea only	7.9[bc]	23.35[c]	378.27[d]
(T2) Urea + HA	7.8[c]	25.69[c]	532.38[c]
(T3) Urea + TSP + MOP + HA	7.7[c]	28.02[c]	651.47[c]
(T4) Urea + TSP + MOP	8.4[ab]	30.36[c]	665.48[b]
(T5) Urea + FA	6.1[e]	112.08[a]	1167.50[a]
(T6) Urea + TSP + MOP + FA	6.2[e]	123.76[a]	1169.84[a]
(T7) Urea + HFA	2.9[f]	53.71[b]	672.48[b]
(T8) Urea + TSP + MOP + HFA	2.8[f]	58.38[b]	546.39[c]
(T9) Urea + humin	-	-	-
(T10) Urea + TSP + MOP + humin	-	-	-
Greenhouse study			
(T0) Soil	5.67[j]	205.48[f]	128.43[k]
(T1) Urea only	6.13[e]	284.87[e]	782.23[i]
(T2) Urea + HA	5.94[g]	221.83[f]	1074.10[g]
(T3) Urea + TSP + MOP + HA	6.53[b]	322.23[d]	1905.36[d]
(T4) Urea + TSP + MOP	6.04[f]	445.99[b]	1625.16[f]
(T5) Urea + FA	5.17[k]	532.38[a]	2066.48[c]
(T6) Urea + TSP + MOP + FA	5.86[h]	294.21[e]	2731.95[a]
(T7) Urea + HFA	5.73[i]	347.92[c]	1832.98[e]
(T8) Urea + TSP + MOP + HFA	6.19[d]	207.82[f]	2419.06[b]
(T9) Urea + humin	6.22[c]	226.50[f]	607.10[j]
(T10) Urea + TSP + MOP + humin	6.63[a]	338.58[cd]	922.33[h]

Different letters within a column indicate significant difference between means using Tukey's test at p = 0.05.

Lower retention of soil pH, exchangeable ammonium and available nitrate resulting from T0 suggest that there was no contribution from soil alone (Table 4). The high values of soil pH, organic carbon, CEC and exchangeable calcium may be due to soil liming. The low pH of HA and FA suggests that they were fully saturated with hydrogen ions during the fractionation process via acidification using 6 N H_2SO_4.

Continuous hydrolysis of urea in T3 caused an increase in soil pH. A high concentration of dissolved ammonia enhanced the fixation of soil exchangeable ammonium (Susilawati et al., 2009b). In a previous laboratory study (Rosliza et al., 2009a, b), the same fertilizer formulation significantly reduced ammonia volatilization by 13% and doubled the retention of exchangeable ammonium compared to the control (Tables 2 and 4). However, soil pH and available nitrate were not significantly different from the control (Table 4). High retention of ammonium in the soil solution suggests that sufficient amount of nitrogen was available for photosynthesis resulting in high DMP of the test crop. Effect of ammonia volatilization reduction due to the soluble salts of K from MOP (Fenn et al., 1982; Rappaport and Axley, 1984), the acidity of H_3PO_4 from TSP (Bock and Kissel, 1998) and the high CEC associated with HA functional groups (Rosliza et al., 2009a, b; Ameera et al., 2009; Regis et al., 2009; Susilawati et al., 2009a; Ahmed et al., 2003, 2006a, b, 2008a; Siva et al., 1999; Fan and Mackenzie, 1993; Stumpe et al., 2003) ensured good retention of soil exchangeable ammonium (Rosliza et al., 2009a, b) and other essential nutrients for good growth of the test crop.

High mobility of fertilizer fractions of the liquid fertilizer applied (Susilawati et al., 2009b) and high exchange capacity (carboxyl groups) of FA (T6) could be the reasons for the observed low soil pH. In a previous laboratory study (Rosliza et al., 2009a, b), a similar fertilizer formulation significantly reduced ammonia volatilization (30%) with the highest retention of soil exchangeable ammonium and available nitrate and lower soil pH (Tables 2 and 4). The carboxyl (COOH) groups which contributes to the exchange capacity of FA is twice that of HA, and at pH 3, protons dissociate (Tan, 2003), which might have enabled FA to retain the highest amount of soil exchangeable ammonium from the fertilizer.

In a previous laboratory study (Rosliza et al., 2009a, b), T8 resulted in remarkable results by controlling 100% of ammonia volatilization with significant retention of soil exchangeable ammonium, available nitrate but very low soil pH compared to the control (Tables 2 and 4). The low pH observed in the laboratory study did not necessarily suggest that the same effect will be obtained when applied to field plants as the effect could be temporary (Ahmed et al., 2008b). Since the soil pH obtained in this pot experiment was optimum for plant growth, it is therefore expected to have good results in future experiments.

The results of T10 with T9 suggest that the improvement in DP was due to TSP and MOP since humin alone (T9) did not significantly improve the dry matter of the test crop (Table 3). T9 also did not improve ammonium retention and availability of soil available nitrate. The high pH of both treatments reflects the low total acidity associated with humin (T10) (Table 4). This can be explained by the low retention of ammonium compared to the control, hence the poor growth of the test crop in the mixture of urea amended with humin. These two fertilizers were added so as to maximize the use of waste in the course of producing the fertilizers of this study. In any future field study, humin is suggested to be applied on top of the soil to control water loss.

For the treatment without TSP and MOP, FA proved to be good because without TSP and MOP (T5), it caused significant retention of ammonium and nitrate and the lowest soil pH due to high total acidity (Table 4). In a previous laboratory study (Rosliza et al., 2009a, b), a similar fertilizer formulation significantly reduced ammonia volatilization (by 23%) with significant retention of soil exchangeable ammonium, available nitrate and lower value of soil pH compared to control (Tables 2 and 4).

From the results, the use of TSP and MOP improved plant total weight and soil exchangeable ammonium and available nitrate retention since the better results for DMP of the test crop was from the mixture of urea, TSP and MOP (T4) (Tables 3 and 4). The mixture of urea and FA (without TSP and MOP) also improved DMP of the test crop suggesting that FA was effective in promoting plant growth (Table 3).

Under laboratory conditions (Rosliza et al., 2009a, b), urea amended with HA had no significant effect in reducing ammonia volatilization, retaining available nitrate and pH compared to the control (Tables 2 and 4). However, this fertilizer formulation was selected for further evaluation under greenhouse conditions due to its ability of doubling the retention of soil exchangeable ammonium (Table 4). High soil ammonium retention was hypothesized to provide better performance in plant growth. However, this fertilizer did not significantly improve dry matter of the test crop but again retained significantly higher soil exchangeable ammonium compared to the control. Soil pH was significantly lower and available nitrate was not significantly different from the control (Table 4). Similar results by Susilawati et al. (2009b) indicated that the inefficiency of HA fertilizers was due to the small amount of HA.

Under laboratory conditions, a similar fertilizer formulation resulted in remarkable results by controlling 100% ammonia volatilization and doubled the retention of soil exchangeable ammonium and available nitrate compared to the control (Tables 2 and 4). However, soil pH was found to be very low under laboratory conditions compared to soil under greenhouse conditions suggesting that the effect of soil pH was temporary (Table 4).

Conclusion

Amending urea with FA and humin increases maize dry matter and urea use efficiency. The findings in this study may only be applicable to similar acid soils. The outcome of this study may contribute to the improvement of urea N use efficiency as well as reducing environmental pollution.

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