

Effects of short-term application of arbuscular mycorrhizal fungi and poultry manure on improvement of soil quality

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ABSTRACT

A field study was conducted to investigate the short-term effect of arbuscular mycorrhizal fungi (AMF) and poultry manure (PM) application on improvements of some selected soil properties compared to NPK chemical fertilizer cropped to maize. PM was applied in tones (t) ha⁻¹ (0, 4, 6, 8, 10 & 12) inoculated with AMF (+AMF) and without AMF (-AMF). Soil samples (0-15 cm) were collected from field according to treatments after maize growth to determine; physical (bulk density, water-stable aggregate [WSA]), chemical (pH, NO₃-N, K, organic carbon [OC] & P) and biological properties (dehydrogenase, urease, phosphatase, AMF spore counts and root colonization [RC]). Results revealed significant improvement in soil properties due to application of poultry manure with and without AMF compared to control and chemical fertilizer. Integration of PM and AMF at 12 t PM ha⁻¹ significantly increased soil pH, NO₃-N, OC, available P, K, % WSA, dehydrogenase, urease and phosphatase activities with reduced bulk density compared to all the treatments. Residual soil NO₃-N, available P & K content at 12 t PM+AMF indicated increment by 11.4% N, 5.8% P, 15.2% K, 25.9% OC, and 21.4% WSA over RD NPK. Spore counts and RC % increased with addition of poultry, the highest recorded at 12 t PM+AMF (17.33±1.202 g⁻¹ soil, 43.00±38.70 %). There was strong positive correlations between RC% and dehydrogenase (R²= 0.854), phosphatases (R²= 0.894), urease (R²= 0.935) activities and WSA (R²= 0.958). Results suggested that application of poultry manure and AMF inoculation could improve soil physical, chemical and biological properties thus could be regarded as a reliable option for maintenance of soil quality and sustainability of crop production.

Key words: Mycorrhiza, chemical fertilizer, enzyme activities, soil quality, poultry manure

INTRODUCTION

Fertilizers are basically added to soil to increase nutrients availability for optimum crop production. Chemical fertilizer has undoubtedly played a major role in food security over the past four decades. Consequently, long-term and any how application has degraded the physical, chemical and biological properties of soil thus, reducing its productive capacity [1]. Yield has become low or stagnant in some regions even with the application of chemical fertilizer which could be related to low soil organic carbon [40,7], poor soil structure [41], and low microbial biomass [12]. Good agronomic practice that would restore and maintain soil quality is crucial to sustainability of agriculture. It is concluded that degraded soil has low nutrients content and soil organic carbon (SOC), poor soil structure and aggregate stability [3], with less microbial biomass [11] especially AMF [28]. Addition of organic C

rich amendments to soil is the initial step to restore, improve and conserve soil quality due to positive influence on soil structure and aggregation, improve soil fertility [44], microbial community and activities [43, 29].

Similar to organic amendments is a growing recognition in application of Arbuscular mycorrhizal fungi (AMF). AMF inoculation is reported as a reliable strategy for reclamation of degraded land and maintenance of soil quality [13,25]. AMF-plant symbiosis can help plant grow in degraded soil by enhancing better plant nutrition [38,27], increase plant resistant to environmental stress, develop stable soil aggregate that could withstand erosion [54], improve nutrients use efficiency of externally added fertilizer [37] and reduce leaching of soil nutrients [23,8] through extension of extraradical hyphae scavenging for nutrients and water beyond the reach of plant root hairs. Several authors have reported the role of AMF in cycling of nutrients and carbon sequestration [58,42] which is a key to sustainable agro-ecosystems. However, the possibility of relying on AMF to bolster crop production depends on management practices that could favour the proliferation of AMF and soil functional quality.

Soil quality indicators depend largely on physical, chemical and biological properties. Aggregate stability, soil porosity, bulk density (physical properties), pH, EC, CEC, N, P, SOC (chemical properties) and biological properties (soil microbial biomass and enzyme activities) were suggested as basal indicators of soil quality [62,15]. Numerous agricultural management practices have affected the physical, chemical and biological properties of soil which created imbalance in soil ecosystems thus affecting its ecological functionality. It is suggested that addition of organic amendments and AMF inoculation might be a reliable option to fertilize crop and improve soil productive quality [4,19]. Previous study revealed that inoculation of AMF amended with empty palm fruit bunch compost have improved soil quality under glasshouse experiment [49]. However, there is less data on effect of poultry manure and AMF inoculation on soil productive quality in the study area. We hypothesized that application of poultry manure and AMF could improve soil quality after maize harvest. This study was conducted to assess short-term effect of AMF and poultry manure on improvement of soil physical (bulk density and aggregate stability) chemical (pH, N, OC, P, & K) and biological properties (phosphatase, dehydrogenase, urease activities, and AMF spore counts) after maize harvest. The outcome of this study could assist us to develop soil nutrients management strategies for sustaining soil quality.

MATERIALS AND METHODS

The experiment was conducted at Kampong Raeh. Located at (1°20'0" North and 110°29'0" E). Having average rainfall of 247 days per annum with mean annual precipitation between 2,500 and 5,000, and a monthly minimum rainfall recorded around June or July but exceeded 100 mm [5]. The temperatures ranges between 23 °C (73 °F) and 33°C (91° F) in the early hours of the morning and during mid-afternoon respectively with heat index reaching 42 °C (108 °F) during dry season due to humidity reaching to about 85%.

The experiment was laid out in a randomized complete block design (RCBD) comprising of 6 levels of poultry manure in tones (t) ha⁻¹ (0, 4, 6, 8, 10 &12) × 2 levels of AM fungi, inoculated (+AMF) and un-inoculated (-AMF) + recommended dose (RD) of NPK chemical fertilizer making, 13 treatment combinations replicated 3 times. The following scheme was used for categorizing the treatment combinations;

Un-inoculated (-AMF)	Inoculated (+AMF)
0 t PM-AMF (Control)	0 t PM+AMF (AMF+ only)
4 t PM-AMF	4 t PM+AMF
6 t PM-AMF	6 t PM+AMF
8 t PM-AMF	8 t PM+AMF
10 t PM-AMF	10 t PM+AMF
12 t PM-AMF	12 t PM+AMF
RD NPK chemical fertilizer (check)	

The gross plot size for the experiment was 10 m × 20 m (200m²). Before treatment application and planting, the initial physicochemical and biological parameters of the soil were analyzed and presented in table 1. A total of 39 plot units, each 2m² sizes were marked according to treatments and replications. Maize seeds were planted after treatment application. Five soil cores from each plot were collected after maize harvest and pooled to form composite samples (0-15 cm) according to treatment plots. Samples were placed inside a sterilized polythene bags and transported to the laboratory in ice box for subsequent analyses. In the laboratory, each soil sample according to treatments were mixed and divided into two subsamples for physic-chemical and microbiological analysis.

Table 1: Pre-plant physical, chemical and biological properties of soils

Characteristics	Values
Soil texture	Silt loam
Bulk density (Mg m ⁻³)	1.36
pH (Soil:H ₂ O 1:5)	5.3
EC (dS m ⁻¹)	0.67
NO ₃ -N (mg kg ⁻¹)	8.7
Available Phosphorus (mg kg ⁻¹)	6.8
Potassium (mg kg ⁻¹)	113.6
Organic C (%)	1.01
Water-stable aggregate _{1-2 mm} (%)	27.64
AMSc*(10g ⁻¹ soil)	2
Dehydrogenase activity (TPF mg g ⁻¹ 24 h ⁻¹)	0.23
Phosphatase activity (PNP mg g ⁻¹ 24 h ⁻¹)	0.12
Urease activity (NH ₄ -N mg g ⁻¹ 24 h ⁻¹)	0.17

AMSc*: Arbuscularmycorrhizal spore count

Collection of data

Soil samples were air dried in laboratory and analyzed for soil bulk density (BD), water-stable soil aggregate (WSA %), soil reaction (pH), organic carbon (OC), available P, NO₃-N, and K. The bulk density was determined on undisturbed soil using steel cylinder (100 cm³) capacity [10]. Water-stable aggregate was determined by wet-sieving method as outlined by Kemper and Rosenau [39] using nest sieve of different diameter (2, 1, 0.5, & 0.25 mm). Macro-aggregates of 1-2mm class size were measured due to their short-term sensitivity to soil treatments. Soil pH was measured using calibrated pH meter in 1:5 (soil: water) ratio suspension, EC was calibrated with EC meter, % OC was determined by loss-on-ignition method as described by Schulte and Hopkins, [56]. Available P was analyzed using Bray No. 2 method [14] and the absorbance measured at 882 nm in a spectrophotometer. Extractable K was assessed using flame photometer as outlined by Jackson, [36], NO₃-N, using UV-VIS spectrophotometer at 420 nm [34].

AMF spores were isolated by wet-sieving and decanting method as outlined by Brundrett, [16]. Isolated spores were counted under stereo binocular microscope using a counter. Root colonization was determined as described by Phillips and Hayman [53].

$$\% \text{ colonization} = \frac{\text{No. of colonized root}}{\text{Total root no.}} \times 100$$

Soil enzyme activities were investigated for dehydrogenase, urease and phosphatase. Dehydrogenase activity (DHA) was determined as outlined by Tabatabai, [59]; to 1g of soil, add 0.2 ml of 3% (w/v) 2, 3, 5-triphenyl tetrazolium chloride solution and 0.5 ml of 1% glucose solution. The mixture was incubated at 30 °C for a day. To the mixture, 10 ml of methanol was added, then refrigerate for 3 hours. Colour intensity was measured in spectrophotometer at 485 nm

Urease (URE) was determined according to Hoffmann and Teicher, [33] procedure, 10% urea solution was added to 10g of soil. The mixture was incubated for 24 hours at 37 °C. Ammonium formed was measured at 578 nm and activity expressed as NH₄-N mg g⁻¹ 24 h⁻¹.

Phosphatase activity was analyzed as described by Tabatabai [59], P-nitrophenyl phosphate disodium (0.115 M) was used as substrate. Briefly, to 5g of soil, 2 ml of 0.5 M sodium acetate buffer at pH 5.5 and 0.5 ml of substrate incubated at 37°C for 90 min. Mixture was allowed to cool at 2°C for 15 min and centrifuge at 4000 rpm for 5 min. Before centrifuging, 0.5 ml of 0.5 M CaCl₂ was added to mixture. P-nitrophenyl formed was detected at 398 nm.

Data analysis

All data collected were subjected to analysis of variance (ANOVA) under general linear model (SPSS version 19), Pearson correlation coefficient in soil parameters were assessed and differences between means were separated using Duncan's LSD at 5%.

RESULTS

Effects of AMF and poultry manure on physical and chemical properties of soil

Results on soil physical and chemical properties as affected by application of AMF and poultry manure after maize harvest are presented in table 2. There was no significant ($p \leq 0.05$) difference in BD between inoculated and un-inoculated plots. However, a decreasing trend was observed with addition of PM in inoculated and un-inoculated plots. Applying 12 t PM+AMF recorded the lowest BD ($1.20 \pm 0.003 \text{ Mg m}^{-3}$) while RD NPK had the highest (1.36 ± 0.003) value that is comparable to 0 t PM-AMF (1.35 ± 0.003).

Table2: Effect of AMF and poultry manure on physical and chemical properties of soil cropped to maize under field condition

Treatment	BD (Mg m^{-3})	WSA (%)	pH (H_2O)	% OC	Av. P	Av. K (mg kg^{-1})	$\text{NO}_3\text{-N}$
Un-inoculated (-AMF)							
0 t PM-AMF (control)	1.35 ^a	27.70 ^a	5.27 ^a	0.97 ^a	5.43 ^a	105.73 ^a	7.23 ^a
4 t PM-AMF	1.30 ^{ac}	27.83 ^a	5.33 ^a	1.12 ^a	6.97 ^b	142.9 ^{3c}	8.80 ^{ab}
6 t PM-AMF	1.23 ^{ab}	27.97 ^a	5.47 ^b	1.13 ^a	7.13 ^b	143.43 ^{cb}	8.97 ^{ab}
8 t PM-AMF	1.23 ^{ab}	28.37 ^{bc}	5.60 ^c	1.15 ^{ab}	7.47 ^{bc}	143.87 ^{cd}	9.17 ^{bc}
10 t PM-AMF	1.23 ^{ab}	29.27 ^d	5.70 ^c	1.19 ^b	7.67 ^{dc}	143.80 ^{cd}	9.50 ^{dc}
12 t PM-AMF	1.22 ^{ad}	29.33 ^d	5.90 ^d	1.40 ^d	8.13 ^{ac}	144.47 ^{bc}	9.80 ^{ac}
Inoculated (+AMF)							
0 t PM+AMF (AMF only)	1.28 ^e	28.23 ^{bc}	5.47 ^b	1.14 ^a	6.87 ^b	143.37 ^{cb}	8.97 ^{ab}
4 t PM+AMF	1.25 ^{bc}	28.50 ^{ac}	5.57 ^b	1.18 ^b	7.27 ^{bc}	143.67 ^{cb}	9.13 ^{bc}
6 t PM+AMF	1.24 ^b	28.70 ^{ac}	5.67 ^c	1.21 ^b	7.47 ^{bc}	143.80 ^{cd}	9.53 ^{dc}
8 t PM+AMF	1.22 ^{ad}	30.33 ^{dc}	5.70 ^c	1.29 ^c	7.67 ^{dc}	144.50 ^{bc}	9.93 ^{ac}
10 t PM+AMF	1.22 ^{ad}	34.17 ^{ae}	5.90 ^d	1.38 ^d	8.00 ^{dc}	145.53 ^{ac}	10.37 ^d
12 t PM+AMF	1.20 ^{ad}	35.00 ^{ed}	6.03 ^d	1.47 ^{bc}	8.33 ^{ac}	145.90 ^{ac}	10.53 ^d
RD NPK	1.36 ^a	27.50 ^a	5.10 ^a	0.98 ^a	6.71 ^b	123.67 ^b	6.80 ^b
Basal soil	1.36	27.64	5.3	1.01	6.8	113.6	8.7

Values are the mean of replicates. Means within same column followed with different superscript are significantly different at $P < 0.05$ according to DMRT

Water-stable aggregate varied significantly between inoculated and un-inoculated plots and increases with addition of poultry manure. RD NPK plots recorded the lowest %WSA (27.50 ± 0.058 %) while 12 t PM+AMF recorded the highest (35.00 ± 0.153 %) that was significantly higher than all the treatments. There was no statistical ($p \leq 0.05$) difference in soil pH of inoculated and un-inoculated plots. However, pH increases with increase in application of PM. The highest pH (6.03 ± 0.088) was recorded at 12 t PM+AMF but value was statistically comparable to 12 t PM-AMF (5.90 ± 0.153). Residual $\text{NO}_3\text{-N}$, K and P concentration and % OC in soil increases with addition of PM in inoculated and un-inoculated plots (Table 2). Applying 12 t PM+AMF had the highest % OC content (1.47 ± 0.037 %) that was incomparable to all the treatments. Residual K & $\text{NO}_3\text{-N}$ was significantly higher in inoculated plots compared to un-inoculated ones. While, there was no significant difference in residual P between inoculated and un-inoculated plots. Highest value was recorded at 12 t PM+AMF ($8.33 \pm 0.233 \text{ mg kg}^{-1}$) with comparable value to 12 t PM-AMF ($8.13 \pm 0.120 \text{ mg kg}^{-1}$). RD NPK recorded the lowest pH and % OC but values were on par with 0 t PM-AMF (Table 2).

Effects of AMF and poultry manure on soil microbiological parameters

Data for soil biological parameters are presented in Figure 1 and Table 3 for mycorrhizal study and soil enzyme activities respectively. Spore counts and RC % increased with addition of poultry manure. The highest spore density and RC % were recorded at 12 t PM+AM. Soil enzyme activities as shown in Table 3 indicated increase in dehydrogenase, acid phosphatases and urease with addition of poultry manure in inoculated and un-inoculated plots. Manure amended plots with AMF (PM+AMF) or without AMF (PM-AMF) recorded higher enzyme activities compared to RD NPK and control plots (0 t PM-AMF) recording the least. The highest dehydrogenase ($0.74 \pm 0.025 \text{ mg TPF g}^{-1} 24 \text{ h}^{-1}$), phosphatase ($1.00 \pm 0.094 \text{ mg PNP g}^{-1} 24 \text{ h}^{-1}$) and urease ($0.76 \pm 0.034 \text{ mg NH}_4 \text{ g}^{-1} 24 \text{ h}^{-1}$) activities were recorded at 12 t PM+AMF and values were statistically incomparable to all the treatments.

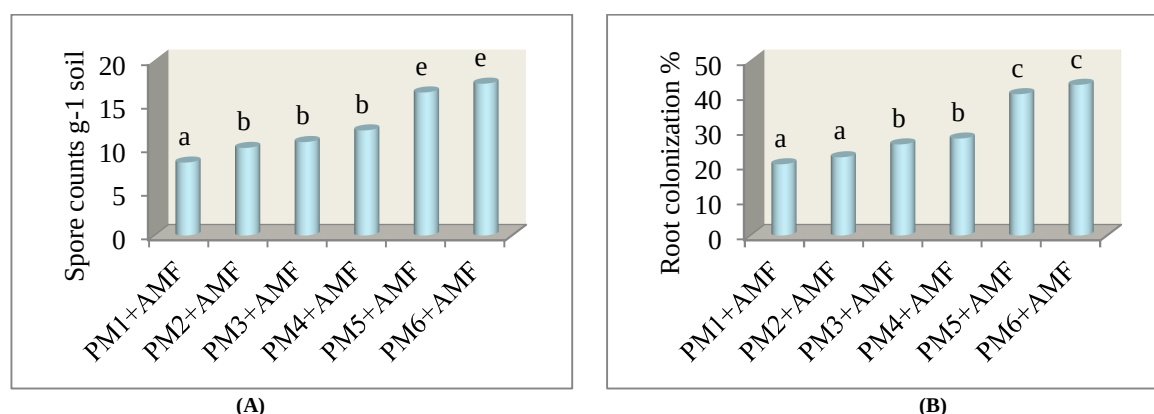


Figure 1: Effect of poultry manure application on AMF spore density (A) and Root colonization (B) in soil after maize harvest

Table 3: Effect of AMF and poultry manure on soil enzyme activities in soil cropped to maize under field condition

Treatments	Dehydrogenase (mg TPF g ⁻¹ 24 h ⁻¹)	Acid phosphatase (mg PNP g ⁻¹ 24 h ⁻¹)	Urease (mg NH ₄ g ⁻¹ 24 h ⁻¹)
Un-inoculated (-AMF)			
0 t PM-AMF (control)	0.28±0.015 ^a	0.16±0.010 ^a	0.21±0.009 ^a
4 t PM-AMF	0.34±0.009 ^b	0.25±0.009 ^a	0.27±0.003 ^b
6 t PM-AMF	0.40±0.012 ^{cd}	0.35±0.018 ^b	0.31±0.012 ^c
8 t PM-AMF	0.46±0.007 ^{ac}	0.41±0.006 ^b	0.37±0.003 ^c
10 t PM-AMF	0.51±0.019 ^{cb}	0.51±0.009 ^d	0.43±0.007 ^d
12 t PM-AMF	0.61±0.009 ^{ad}	0.55±0.009 ^d	0.49±0.003 ^{ac}
Inoculated (+AMF)			
0 t PM+AMF	0.33±0.009 ^b	0.37±0.003 ^b	0.26±0.007 ^b
4 t PM+AMF	0.49±0.003 ^{cb}	0.43±0.012 ^b	0.32±0.007 ^c
6 t PM+AMF	0.55±0.015 ^c	0.53±0.026 ^d	0.44±0.010 ^d
8 t PM+AMF	0.62±0.018 ^{ad}	0.65±0.017 ^{bc}	0.55±0.023 ^{ac}
10 t PM+AMF	0.68±0.010 ^{cc}	0.75±0.012 ^{bc}	0.62±0.021 ^{dc}
12 t PM+AMF	0.74±0.025 ^{bd}	1.00±0.094 ^{dc}	0.76±0.034 ^{cb}
RD NPK	0.32±0.015 ^b	0.17±0.003 ^a	0.25±0.003 ^b

Values are the mean of replicates with mean SE±. Means within same column followed with different alphabet are significantly different at $P < 0.05$ according to DMRT

DISCUSSION

The pre-plant residual soil $\text{NO}_3\text{-N}$ (8.7 mg kg^{-1}) and available PO_4 (6.8 mg kg^{-1}) were below critical value for crop growth [32], with low native AMF spore count ($2 \text{ spores } 10 \text{ g}^{-1} \text{ soil}$), as such, field was not fumigated. Results indicated improvements in physical (bulk density, water-stable aggregate), chemical (K, N, % OC, P and pH) and biological properties (AMF spore density, dehydrogenase, urease and phosphatase activities) of soil due to short-term application of poultry manure and AMF. The present study confirmed the overall benefit of poultry manure application in improving soil quality. It is suggested that organic amendments when incorporated into soil could increase soil organic carbon, decrease bulk density, improve availability of soil macro- and micronutrients [67] and lower soil acidity [61, 46]. As recorded in this study reference have shown high nutrients accumulation and organic matter content in soil from short or long-term application of organic amendments compared to chemical fertilizer [20]. Tambone et al. [60] reported increase in total organic carbon (TOC), N, P and pH from short-term application of compost after maize harvest. Low soil pH due to application of chemical fertilizer as recorded in this study was reported [48] and this could be attributed to the acidifying effect of urea-N fertilizer [6]. The increased in soil pH of inoculated plots and un-inoculated ones over RD NPK (Table 2) could be due to provision of some basic ions in the manure to the soil [17]. It is suggested that short-term and long-term application of manure could reduce acidity in soil due to Ca and Mg contained in the manure. Exudates produced by AMF hyphae could also be an influence to increase in pH of the inoculated plots [64]. In addition, Celik et al. [21] reported lower bulk density and higher organic matter content in soils receiving manure, compost and integration of AMF and compost compared to soil treated with chemical fertilizer, in a long-term study with winter wheat and maize rotation. Low degree of WSA % recorded in RD NPK and control could be attributed to low organic carbon. It is assumed that organic matter binds mineral particles of soil thereby contributing to aggregate stability. Celick et al. [21] confirmed superiority in soil aggregate stability with compost along with AMF compared to compost alone and mineral fertilizer. They

concluded it to low content of organic matter in the soil. Low dehydrogenase, urease and phosphatase activities observed in plot treated with RD NPK were revealed [22,63,45,9]. Conversely, higher enzymatic activities observed in AMF plots are in conformity with some researchers [65]. The application of PM has fostered AMF sporulation compared to inoculated plots without PM amendment (0 t PM+AMF). This could be attributed to improve soil aeration from reduction in bulk density caused by application of manure thus, provided ideal condition for mycorrhizal proliferation [27]. Groanker and Sreenivasa, [31] and Douds et al. [26] reported mycorrhizal development in soil amended with lower C: N such as animal manure. Calvet et al. [18] and Albertsen et al. [2] concluded that addition of organic amendments might improve soil fertility and bolster AMF sporulation.

Consistent results have shown huge role of organic amendments in enhancing microbial communities by adding SOC which benefit the microbial biomass, diversity and activity especially, AMF community [55,52,50,30]. This could be reflected on the increase in residual soil nutrient concentration as microorganisms decompose and utilize C from amended soils [35,66]. Several researchers have reported residual accumulation of $\text{NO}_3\text{-N}$, P and increased pH after long-term application of poultry manure [47,68] due to high content of N, P, K and other nutrient elements in the manure [24,51].

CONCLUSION

Short-term application of poultry manure with and without AMF plots resulted in improving the physical (bulk density, water-stable aggregates) chemical (pH, N, P, OC, & K) and biological (DHA, URE & phosphatase activities) soil properties compared to chemical fertilizer. Poultry manure was effective in increasing AMF spore density in inoculated plots. The integration of poultry manure and AMF at 12 t PM+AMF proved to be most effective in increasing soil pH, OC, WSA and residual accumulation of $\text{NO}_3\text{-N}$, $\text{PO}_4\text{-P}$, & K for plant use over all treatments. The results also indicated higher dehydrogenase, phosphatase and urease activities at 12 t PM+AMF. It could be concluded that integration of AMF and poultry could be a management strategy to improvement of soil productive quality. Thus, could be recommended to farmers and long-term experiment should be conducted to provide better understanding since longer time allows more degradation of added organic amendment.

Acknowledgments

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