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Soil Biochemical Changes Induced by Poultry Litter Application and Conservation Tillage under Cotton Production Systems

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Abstract: Problems arising from conventional tillage (CT) systems (such as soil erosion, decrease of organic matter, environmental damage *etc.*) have led many farmers to the adoption of no-till (NT) systems that are more effective in improving soil physical, chemical and microbial properties. Results from this study clearly indicated that NT, mulch tillage (MT), and winter rye cover cropping systems increased the activity of phosphatase, β-glucosidase and arylsulfatase at a 0–10 cm soil depth but decreased the activity of these enzymes at 10–20 cm. The increase in enzyme activity was a good indicator of intensive soil microbial activity in different soil management practices. Poultry litter (PL) application under NT, MT, and rye cropping system could be considered as effective management practices due to the improvement in carbon (C) content and the biochemical quality at the soil surface. The activities of the studied enzymes were highly correlated with soil total nitrogen (STN) soil organic carbon (SOC) at the 0–10 cm soil depth, except for acid phosphatase where no correlation was observed. This study revealed that agricultural practices such as tillage, PL, and cover crop cropping system have a noticeable positive effect on soil biochemical activities under cotton production.

1. Introduction

Deterioration of soil quality in southeastern USA (the most productive area in agriculture) has been attributed to soil erosion associated with an increase of conventional monoculture practices. In Alabama, eroded soils under conventional cotton production average about 25 t ha^{-1} yr^{-1} [1]. Nevertheless, farmers continue to use intensive conventional tillage (CT) for continuous cotton production in these areas. Conventional tillage has the potential to cause loss of soil organic matter (SOM) and soil nutrients content leading to a decrease in biological activity. To sustain cotton production in the region, efforts have focused on using a conservative system such as no-till (NT).

NT practices are known to have advantages over CT with respect to soil chemical, physical and biological properties mainly in the surface layer [2]. Soil microbes are sensitive to changes in soil from different management practices and can be used to predict the impact of management practices on soil quality. [3,4] stated that conventional agricultural practices inhibit the activity and function of soil microbes with insecticide or/and fungicide application. Thus, studies found that soil microbial populations and enzyme activities involved in carbon (C), nitrogen (N), phosphorus (P) and sulfur (S) cycling were greater with NT [5,6]. Because NT systems affect nutrient mineralization and microbial biomass, this system provides a more favorable habitat for soil microorganisms [7]. To better understand C, N and other nutrient-cycling, it is important to gain understanding of microbial communities and related processes [8]. Results from previous studies have shown that enzyme activities are often used as indices of microbial activity that provide a useful tool to monitor the effects of soil management on long-term soil quality [9].

Apart from microorganisms, plants secrete enzymes to catalyze the hydrolysis of phytate [10,11]. For example, cowpea density significantly increased both acid and alkaline phosphatase activity in cowpea rhizosphere, resulting in improvement of P nutrition, plant growth and yield [12]. Acid phosphatase secretion from plant roots has been reported to increase under P deficiency and differ within and between plant species [13]. Plant roots produce phosphatase in soils under P stress conditions to enhance the solubilization and remobilization of phosphate [13,14]. Glucosidase, arylsulfatase and phosphatase were found to be distinctly higher under the NT system [15], and were key enzymes involved in mineralization of organic C, P, and S in soil.

Phosphatase has the potential to directly and indirectly influence phosphorus transformation by participating in chemical reaction and influencing microbial processes that drive P solubilization [16]. Phosphatase was shown to hydrolyze the carbon-phosphorus ester bond during the mineralization phase of soil organic phosphorus [17]. Some bacteria and fungi are able to enhance the availability of phosphorus to crops through biochemical mechanisms such as, solubilizing inorganic phosphorus by chelation or acidification, mineralizing organic phosphorus by producing extracellular phosphatases or phytases [18].

Other enzymes, such as glycosidases play an important role in the degradation of carbohydrates and provide energy for microorganisms in soil. Among the glycosidases, β-glucosidase is the most abundant in soil. It catalyzes the hydrolysis of β-glucopyranoside, involved in hydrolysis of cellulose to glucose, the source of energy for organisms [19]. Arylsulfatase activity is used to investigate organic S mineralization and can be an indicator of the presence of fungi and bacteria in soil [20].

Previous studies demonstrated that the activity of enzymes such as L-asparaginase, L-glutaminase, and urease were significantly greater under no-till systems than under conventional systems [21,22]. The amount of activity depends on many factors (soil management system, organic matter content, soil texture, soil nutrient composition). The transition from CT to NT system increases microbial and enzyme activities near the soil surface [23]. From the perspective of sustainable production, it is important to understand the dynamics of microorganisms in soil under different management systems. Relatively little is known of the impact of CT *versus* NT on soil microbial communities and soil enzyme activities under cotton production. Therefore, the aim of this study was to investigate the effect of different agricultural management practices [NT, mulch-till (MT), or CT] on soil biological activity in cotton fields under poultry litter (PL) and N fertilizer application. Three key enzymes were chosen to investigate in this study: phosphatases involved in P cycle, β-glucosidase in C cycle, and arylsulfatase in S cycle.

2. Materials and Methods

An experiment was carried out at the Alabama Agricultural Experiment Station, Belle Mina, AL, USA situated at 34°41' N, 86°52' W on crop rotations with tillage treatments including NT, MT and CT systems. All the treatments are listed in Table 1 [24,25].

Table 1. List of treatments used in the cotton field, Belle Mina, AL, USA.

No.	Tillage system	Cropping system	N source	N rate kg ha^{-1}
1 (CTR)	Conventional till	Cotton-rye	None	0
2 (CTAN)	Conventional till	Cotton-fallow	Ammonium nitrate	100
3 (NTAN)	No-till	Cotton-fallow	Ammonium nitrate	100
4 (CTRAN)	Conventional till	Cotton-rye	Ammonium nitrate	100
5 (CTRP)	Conventional till	Cotton-rye	Poultry litter	100
6 (MTRAN)	Mulch-till	Cotton-rye	Ammonium nitrate	100
7 (MTRP)	Mulch-till	Cotton-rye	Poultry litter	100
8 (NTRAN)	No-till	Cotton-rye	Ammonium nitrate	100
9 (NTRP)	No-till	Cotton-rye	Poultry litter	100
10 (NT)	No-till	Cotton-fallow	None	0
11 (NTRPP)	No-till	Cotton-rye	Poultry litter	200
12 (BF)	None	Bare fallow	None	0

The soil is a Decatur silt loam (Clayey, kaolinitic thermic, Typic Paleudults) with a mean particle distribution of 15% sand, 58% silt, and 26% clay. The initial soil pH of 6.2 was measured in a 1:2 (weight/volume) soil/water mixture using a combination electrode. The experimental design was a randomized complete block with an incomplete factorial treatment arrangement. The treatments included three tillage systems (NT, MT and CT), two cropping systems [cotton-without rye (cotton-fallow),

and cotton with rye (cotton-rye)], and two sources of nitrogen [ammonium nitrate (NH_4NO_3) and PL] in four replications. Plots were 8 m wide and 9 m long, which resulted in 8 rows of cotton spaced 1 m apart. The total amounts PL and NH_4NO_3 were broadcasted by hand one day before planting and incorporated to a depth of 5 cm at the rate of 100 for NH_4NO_3 and 100 or 200 kg N ha^{-1} for PL. The ammonium nitrate application rate (100 kg N ha^{-1}) was applied based on the Alabama A&M University Extension Service recommendation for cotton production in the region [26]. Poultry litter rates were applied based on changes in N release associated with the high concentration of N in PL. The 200 kg N ha^{-1} PL treatment was only applied to NT plots. Nevertheless, only the 100 kg N ha^{-1} PL treatment was used in this study. Triple superphosphate and muriate of potash were both applied at rate of 67 kg ha^{-1} on all plots including PL plots to nullify the effects of P and K from PL. Glyphosate was used for weed control in NT and MT systems. The typical conservational tillage systems used in the region included moldboard, chisel, and disk plowing were used in the study. The working depth for moldboard and chisel plowings was about 15 cm, while the depth of the disk plow was 10–15 cm before cotton seeding. Tillage operations occurred two times a year, moldboard plowing in fall after crop harvest, and disking and leveling with a field cultivator in spring before summer crop planting. Mulch till included only chisel plowing to partially incorporate crop residues into soil before planting. Smooth seed beds were prepared in both CT and MT plots using a rotary field cultivator. Under no-till plots, soil was disturbed only to the extent of opening furrows for seeding purpose. Poultry litter and NH_4NO_3 were incorporated into soil in conventional and mulch tillage plots, and left unincorporated in no-tillage plots.

2.1. Soil Sampling and Enzyme Analyses

Soil samples for this study were randomly collected from each plot at two different depths (0–10 cm and 10–20 cm), 4 weeks after cotton was harvested, and a day after the first rainfall following the cold and dry January and February months. Sterile polypropylene bags were filled with field-moist soil samples, and stored at 4 °C for analysis. Cumulative rainfall from January to April 2006 was only 6 mm. The top 10 cm and 20 cm soil temperatures were consistently 17 °C and 14 °C, respectively (Belle Mina Research Station Survey, 2006). The activities of phosphomonoesterase (acid and alkaline phosphatases) and phosphodiesterase were assayed using air-dried soil as previously described by [27]. Phosphomonoesterase activity was assayed by incubating 1 g of air-dried soil sample at 37 °C for 60 min, with 0.2 mL of toluene and 4 mL of modified universal buffer (MUB) buffer at pH 6.5 for acid and pH 11.0 for alkaline phosphatases. The reaction was terminated by adding 1 mL of 0.5 M $CaCl_2$ and 4 mL of 0.5 M NaOH to prevent dispersion of humic substances. The assay of phosphodiesterase activity used a method similar to that of phosphomonoesterase with the only difference being 4 mL of THAM [Tris (hydroxymethyl) aminomethane] buffer at pH 8. This reaction was terminated by adding 1 mL of 0.5 M $CaCl_2$ and 4 mL of THAM-NaOH to prevent dispersion of humic substances. The mixture was centrifuged at 15,000*g* for 10 min, and the product of the reaction *p*-nitrophenol phosphate (PNP) with phosphomonoesterase and bis-*p*-nitrophenyl phosphate (bis-PNP) with phosphodiesterase were measured at 410 nm using a spectrophotometer. The arysulfatase and β-glucosidase activities were measured the same way as phosphomonoesterase activity, buffered respectively at (pH 5.8) with potassium *p*-nitrophenyl sulfate, and buffered at (pH 6.0) with

p-nitrophenyl glycoside solution [28]. Controls were performed for all enzymes assayed by adding substrate immediately after the addition of $CaCl_2$ and NaOH incubation prior to analysis.

2.2. Statistical Analyses

Analysis of variance, contrast comparisons and mean separation was conducted using the GLM procedure of the statistical analysis software, SAS [29], to assess the effects of tillage, cropping systems, and N sources as related to soil depth. Because treatments were laid out in an incomplete factorial arrangement, selected treatments were used for this study. The following treatment CTAN, NTAN, CTRAN**,** and NTRAN (Table 1) were used to determine the effect of tillage, cropping system, and tillage × cropping system interaction on selected enzyme activities. Similarly, CTRAN, CTRP, MTRAN, MTRP, NTRAN, and NTRP treatments were used to evaluate the effect of N sources and tillage × N source interaction. Least significant difference analysis (LSD) was preformed to permit means separation.

3. Results

3.1. Soil Enzyme Activities

Acid phosphatase and phospodiesterase activities were significantly ($p < 0.05$) affected by the tillage systems and soil sampling depths at 10–20 cm and 0–10 cm respectively (Table 2).

Under the different tillage systems, acid phosphatase activity was almost three times higher than alkaline phosphatase activity, and 10 times higher than phospodiesterase phosphatase (Figure 1).

Figure 1. Effect of tillage systems on phosphatase, β-glucosidase, and arylsufatase at two different soil depths. Different letters indicate significant differences between tillage for each enzyme activities.
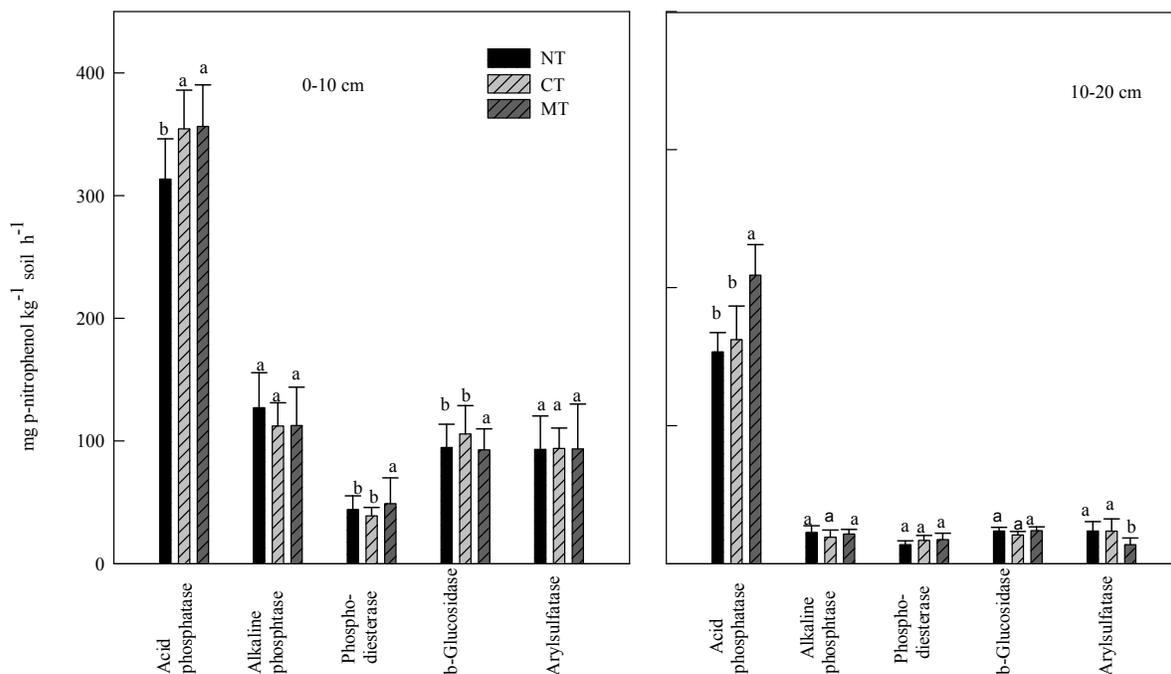
Table 2. Effect of tillage, cropping system, and N sources on selected enzyme activities at 0–20 cm soil depth.

	Acid phoshatase		Alkaline phosphatase		Phospho-diesterase		Glucosidase		Arylsulfatase	
Depth →	1	2	1	2	1	2	1	2	1	2
P value										
Tillage and N source on (Treatment 4, 5, 6, 7, 8, and 9 ‡)										
Tillage (T)	ns	***	ns	ns	*	ns	ns	ns	ns	ns
N Sources (N)	ns	ns	**	ns	ns	ns	***	ns	**	ns
T × N	ns	**	*	ns	ns	ns	***	*	ns	ns
Tillage and cropping systems on (Treatment (2, 3, 4, and 8)										
Tillage (T)	ns	***	ns	ns	*	ns	ns	ns	ns	ns
Cropping (C)	ns	ns	ns	ns	*	ns	***	*	***	*
T × C	ns	ns	ns	ns	**	*	***	ns	*	*

Depth: 1 = 0–10 cm; 2 = 10–20 cm; ‡ See Table 1 for treatments description; * Significant at $P \leq 0.05$; ** Significant at $P \leq 0.01$; *** Significant at $P \leq 0.001$.

As a result, the activity of phospodiesterase appears as the least important phosphatase within the phosphatase activity pools in soils. Statistical differences in tillage were observed with glucosidase activities at 0–10 cm soil depth and arylsulfatase activities at 10–20 cm depth.

3.2. Effect of Rye Cover Crops on Soil Enzyme Activities

Figure 2 showed the enzyme activities under two cropping systems (rye and no rye). At the 0–10 cm soil depth, the activity of the acid phosphatase was 344.47 and 301.84 mg PNP kg^{-1} soil h^{-1}, the alkaline phosphatase was 99.0 and 97.20 mg PNP kg^{-1} soil h^{-1} under rye and no rye application systems respectively, while that of the phosphodiesterase was 42.0 and 27.96 mg bis-PNP kg^{-1} soil h^{-1} under rye and no rye application. There were no significant differences between cover crop rye and no rye treatments for phosphomonoesterase activity at 0–10 cm and no cover crop difference for any enzyme activity at 10–20 cm. The cover crop (rye) application showed higher β-glucosidase activity at 0–10 cm soil depth than with the no-rye application at the same depth when comparing cropping system. This suggests that rye application increases the activity of β-glucosidase, probably due to the effect of cover crop residues left in the upper plow layer of soil. In addition, arylsulfatase activity and phosphodiestrase was significantly affected by cover crop (rye) although no difference was observed with depth.

3.3. Effect of Poultry Litter and NH_4NO_3 Application on Soil Enzyme Activities

No significant treatment differences were found under PL application, however, all the enzyme activities increased in organically fertilized plots at 0–10 cm soil sampling depth (Figure 3) The application of PL may have improved organic matter status in soils, which in turn enhanced the enzyme activities [30].

Figure 2. Effect of cover crop (rye) on selected enzyme activities at two soil sampling depths. The bars with different letters within enzyme activity are significantly different ($P < 0.05$).
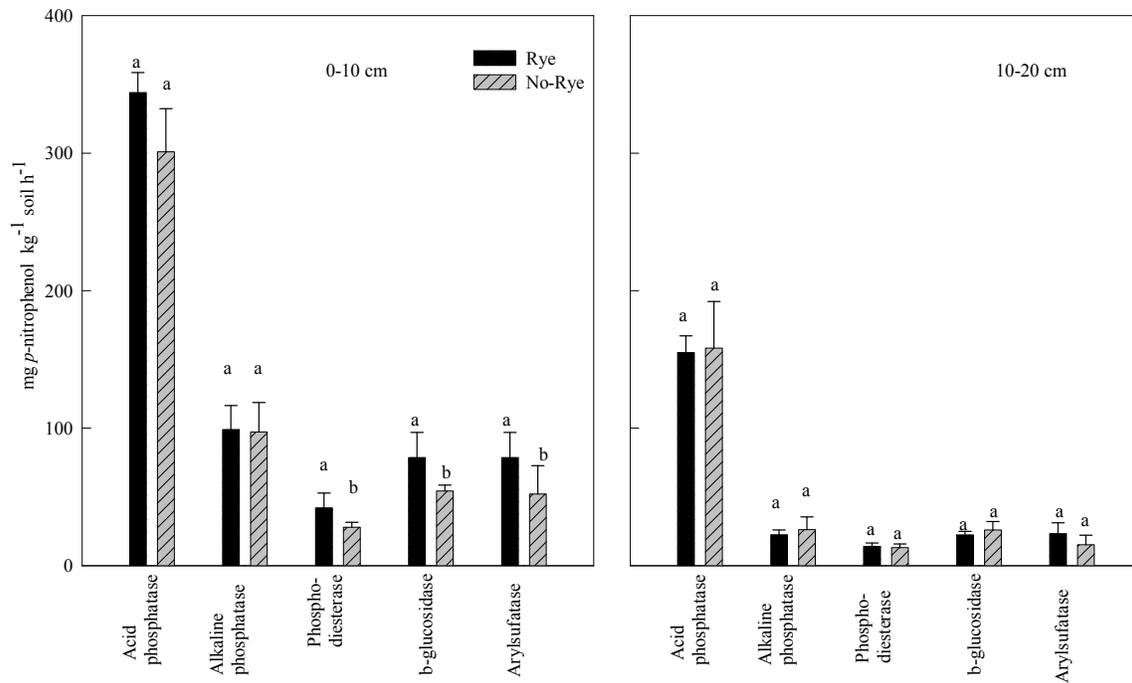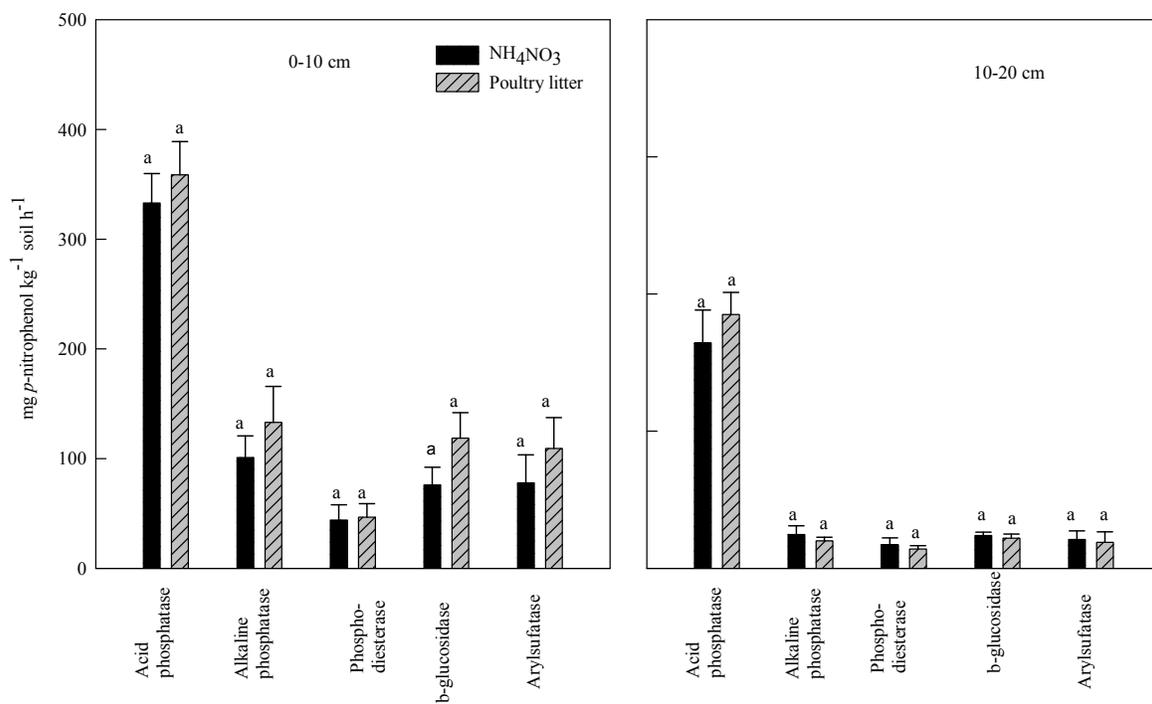

Figure 3. Effect of poultry litter and ammonium nitrate on average values of selected enzyme activities at two soil sampling depths. The bars with different letters within enzyme activity are significantly different ($P < 0.05$).

3.4. Correlation between Soil Enzyme Activities and Organic Carbon, and Total Nitrogen

All enzymatic activities were highly correlated with soil organic C or soil organic N, except for acid phophatase where no correlation was observed. Alkaline and phosphodiesterase phosphatases, β-glucosidase, and arylsulfatase activities (Table 3) significantly correlated with soil organic C ($r = 0.58***, 0.50***, 0.58***$, and $0.66***$ respectively) and soil total N ($r = 0.47**, 0.46**, 0.39**$, and $0.48**$ respectively). Other authors have reported similar results with soil organic C [23] and soil total N [31]. However, acid phosphatase was only correlated to N at 10–20 cm depth.

Table 3. Correlation coefficients between enzyme activities and selected soil properties.

	Acid Phosphatase		Alkaline Phosphatase		Phospho-Diesterase		Arylsulfatase		Organic C		Total N	
Depth ➡	1	2	1	2	1	2	1	2	1	2	1	2
β-glucosidase	0.30 *	0.10	0.55 ***	0.30 *	0.46 **	0.04	0.75 ***	−0.003	0.58 ***	0.16	0.39 **	0.15
Acid Phosphatase			−0.009	−0.04	0.06	0.01	0.21	−0.25	0.19	0.17	0.10	0.30 *
Alkaline Phosphatase					0.59 ***	0.08	0.68 ***	0.06	0.58 ***	0.23	0.47 **	0.16
Phospho-Diesterase							0.62 ***	0.21	0.50 ***	0.36 **	0.46 **	0.03
Arylsulfatase									0.66 ***	0.23	0.48 ***	0.26

Depth: 1 = 0–10 cm; 2 = 10–20 cm; * Significant at $P ≤ 0.05$; ** Significant at $P ≤ 0.01$; *** Significant at $P ≤ 0.001$.

The GLM test showed that tillage and tillage × cropping significantly increased β-glucosidase activities at the 0–10 cm depth, which also occurred when tillage and N source were combined.

4. Discussion and Conclusions

The results from this study show low phosphatase activities in both soil depths as compared to those reported by [26] in an Australian forest. The build-up of organic matter content under NT practices, not only improve soil structure and water retention but also serves as a source for plant nutrients and substrate for soil microbes. [1, 22] reported that phosphatase activities were more than twice as high in the surface layer in the NT practice than in the CT practice. Phosphatases are adaptive enzymes and the intensity of secretion by microorganisms or plant roots depend on their need for phosphates [32]. The lower phosphatase activities in NH_4NO_3 than PL were likely due to high P availability from high rate of PL application. The application of PL based on N concentration may have increased soil to the point that it reduced the activity of microorganisms responsible for phosphatase production. In previous studies greater activity of phosphomonoesterases was observed in acidic soil rich in fungi [33,34]. In contrast, phosphodiesterase activity was greater in neutral soils with enzymes synthesis mainly by bacteria or actinomycetes [35]. In this study initial soil pH of 6.2 may have negatively affected the activity of phosphomonoesterase. The higher acid and phosphodiesterase phosphatases in mulch treated plots compared to NT and CT could be the result of a flush of organic residue on the soil surface. Incorporation of crop residues into soil enhances enzyme production [36]. This indicates that phosphatase could be used as a good health predictor in CT and NT plots. In our

study, alkaline phosphatase was positively correlated to SOC and N at 0–10 cm soil depth. It is believed that alkaline phosphatase is important for determining the relative activity of microbial population in soils.

β-Glucosidase enzymes are widely distributed in nature, and are known to play an important role in the C cycle [37]. The highest enzyme activities were found in mulch plots, followed by NT and CT plots. This finding indicates a strong dependence of β-glucosidase on SOC. The organic residues from rye and mulch were an adequate substrate for stimulating microbial activity and β-glucosidase liberation. The combination of PL and rye residues may have promoted β-glucosidase activity [38]. β-glucosidase enzyme is known to catalyze the hydrolysis of cellulose, an important energy source for soil microorganisms. The activity of the β-glucosidase was closely associated with SOC (0.80, $P \leq 0.05$) (Table 2). This correlation was due to the involvement of β-glucosidase in the C cycle related to soil organic matter recycling.

Arylsulfatases and alkaline phosphatase mostly originate from microbes such as bacteria and fungi that hydrolyze sulfate ester in soil [23]. Arylsulfatase catalyzes the hydrolysis of organic sulfate, and the differences observed when comparing cropping systems (rye and no-rye) are due to the changing form of S contained in the decomposed surface residue on the NT systems. The activity of arylsulfatase increases with N fertilization and is used as an indicator of microbial activities. Higher amounts of arysulfatases were observed under PL application and decreased with depth (Figure 3). No major differences in arylsulfatase were observed among tillage systems or N sources. The lack of significant difference between tillage systems suggested that tillage operations probably had little influence on microbial activities at 0–10 cm depth. The significant correlation of the arylsulfatase activity with SOC at 0–10 cm depth reflected the connection between enzyme substrates and the carbon molecules [27]. These results were in agreement with those obtained by [39], where C and N mineralization was an indicator of biologically-active organic matter. The relationship with SOC suggested that C addition through rye residue stimulated microbial activity, and could thereby enhance the overall assimilation of C by microorganisms [16]. Organic matter plays an important role in maintaining soil enzymes in active forms. Soil organic amendments could enhance soil enzyme activities by increasing soil organic matter and therefore, the microbial biomass [40].

The enzymes' activities were inter-correlated with r value up to 0.75 and highly correlated with SOC and total N. This indicates that enzyme activities responded positively to the soil management. On soil surfaces, reported SOC trends are generally in the order NT > MT > CT (see USDA practice standard 329 for NT; practice Standard 345 for Mulch-till). This implies higher carbon sequestration rates for NT compared to MT, and also for MT as compared to CT. Therefore, factors such as enzyme activities can be used as indicators of short-term biochemical change, since they integrate information on microbial status in soil. Because these activities were sensitive to tillage practices and cropping systems they can be used to delineate management activities under cotton production.

References

1.   Nyakatawa, E.Z.; Reddy, K.C.; Sistani, K.R. Tillage, cover cropping, and poultry litter effects on selected soil chemistry properties. *Soil Tillage Res*. **2001**, *58*, 69–74.

2. Mikanova, O.; Javurek, M.; Simon, T.; Friedlova, M.; Vach, M. The effect of tillage systems on some microbial characteristics. *Soil Tillage Res*. **2009**, *105*, 72–76.

3. Acosta-Martinez, V.; Mikha, M.M.; Vigil, M.F. Microbial communities and enzyme activities in soils under alternative crop rotations compared to wheat–fallow for the central great plains. *Appl. Soil Ecol.* **2007**, *37*, 41–52.

4. Liu, B.; Tu, C.; Hu, S.; Gumpertz, M.; Ristaino, J.B. Effect of organic, sustainable, and conventional management strategies in grower fields on soil physical, chemical, and biological factors and the incidence of Southern blight. *Appl. Soil Ecol*. **2007**, *37*, 202–214.

5. Alvear, N.; Rosas, A.; Rouanet, J.L.; Borie, F. Effect of three soil tillage systems on some biological activities in an ultisols from southern Chile. *Soil Tillage Res*. **2005**, *8*, 195–202.

6. Omidi, H.; Tahmasebi, Z.; Torabi, H.; Miransari, M. Soil enzymatic activities and available P and Zn as affected by tillage practices, canola (*Brassica napus* L.) cultivars and planting dates. *Eur. J. Soil Biol.* **2008**, *44*, 443–450.

7. Balota, E.L.; Colozzi-Filho, A.; Andrade, D.S.; Dick, R.P. Long-term tillage and crop rotation effects on microbial biomass and C and N mineralization in a Brazilian Oxisol. *Soil Tillage Res*. **2004**, *77*, 137–145.

8. Ingram, L.J.; Stahl, P.D.; Schuman, G.E.; Buyer, J.S.; Vance, G.F.; Ganjegunte, G.K.; Welker, J.M.; Derner, J.D. Grazing impacts on soil carbon and microbial communities in a mixed-grass ecosystem. *Soil Sci. Soc. Am. J.* **2008**, *72*, 939–948.

9. Dick, R.P.; Doran, J.W.; Coleman, D.C.; Bezdicek, D.F.; Stewart, B.A. Soil Enzyme Activities as Indicators of Soil Quality. In *Defining Soil Quality for a Sustainable Environment*; Soil Science Society of America (SSSA): Madison, WI, USA, 1994; pp. 107–124.

10. Dodd, J.C.; Burton, C.C.; Burns, R.G.; Jeffries, P. Phosphatase activity associated with the roots and the rhizosphere of plants infected with vesicular-arbuscular mycorrhizal fungi. *New Phytol.* **1987**, *107*, 163–172.

11. Tazisong, I.A.; Senwo, Z.N.; Taylor, R.W.; He, Z. Hydrolysis of organic phosphates by commercially available phytases: Biocatalytic potentials and effects of ions on their enzymatic activities. *J. Food Agric. Environ*. **2008**, *6*, 500–505.

12. Makoi, J.H.; Chimphango, S.B.M.; Dakora, F.D. Elevated levels of acid and alkaline phosphatase activity in roots and rhizosphere of cowpeas (*Vigna unguiculata* L.Walp) genotypes grown in mixed culture and at different densities with sorghum (*Sorghum bicolor* L.). *Crop Pasture Sci.* **2010**, *6*, 279–289.

13. Mudge, S.R.; Rac, A.L.; Diatloff, E.; Smith, F.W. Expression analysis suggests novel roles for members of Pht1 family of phosphate transporters in Arbidopsis. *Plant J*. **2002**, *31*, 341–353.

14. Richardson, A.E.; Hadobas, P.A.; Hayes, J.E. Phosphomonoesterase and phytase activities of wheat (*Triticum aestivum* L.) roots and utilization of organic phosphorus substrates by seedlings grown in sterile conditions. *Plant Cell Environ.* **2000**, *23*, 397–405.

15. Bergstorm, D.W.; Monreal, C.M.; King, D.J. Sensitivity of soil enzyme activities to conservation practices. *Soil Sci. Soc. Am. J*. **1998**, *62*, 1286–1295.

16. Bunemann, E.K.; Steinebrunner, F.; Frossard, P.C.; Oberson, A. Phosphorus dynamics in a highly weathered soil as revealed by isotopic labeling techniques. *Soil Sci. Soc. Am. J*. **2004**, *68*, 1645–1655.

17. Sharpley, A.N.; Rekolainen, S. Phosphorus in Agriculture and Its Environmental Implications. In *Phosphorus Loss from Soil to Water*; Tunney, H., Carton, O.T., Brookes, P.C., Johnston, A.E., Eds.; Commonwealth Agricultural Bureaux (CABI): Wallingford, UK, 1997; pp. 1–54.

18. Jakobsen, I.; Leggett, M.E.; Richardson, A.E. Rhizosphere Microorganisms and Plant Phosphorus Uptake. In *Phosphorus. Agriculture and the Environment*; Sims, J.T., Sharpley, A.N., Eds.; American Society of Agronomy (ASA) Crop Science Society of America (CSSA) Soil Science Society of America(SSSA): Madison, WI, USA, 2005; pp. 437–492.

19. Martinez, C.E.; Tabatabai, M.A. Decomposition of biotechnology by products in soils. *J. Environ. Qual*. **1997**, *26*, 625–632.

20. Acosta-Martinez, V.; Upchurch, D.R.; Schubert, A.M.; Porter, D.; Wheeler, T. Early impacts of cotton and peanut cropping systems on selected soil chemical, physical, microbiological and biochemical properties. *Biol. Fertil. Soils* **2004**, *40*, 44–54.

21. Deng, S.P.; Tabatabai, M.A. Effect of tillage and residue management on enzyme activities in soils: I. amidohydrolases. *Biol. Fertil. Soils* **1996**, *22*, 202–207.

22. Deng, S.P.; Tabatabai, M.A. Effect of tillage and residue management on enzyme activities in soils phosphatase and arysulfatase. *Biol. Fertil. Soils* **1997**, *24*, 141–146.

23. Acosta-Martinez, V.; Tabatabai, M.A. Enzyme activities of a lime agricultural soil. *Biol. Fertil. Soils* **2000**, *31*, 85–91.

24. Sainju, U.M.; Senwo, Z.N.; Nyakatawa, E.Z.; Tazisong, I.A.; Reddy, C.K. Soil carbon and nitrogen sequestration as affected by long-term tillage cropping systems, and nitrogen fertilizer sources. *Agric. Ecosyst. Environ.* **2008**, *127*, 234–240.

25. Nyakatawa, E.Z.; Reddy, K.C.; Mays, D.A. Tillage, cover cropping, and poultry litter effects on cotton: II. Growth and yield parameters. *Agron. J.* **2000**, *92*, 1000–1007.

26. Adams, M.A. Phosphatase activity and phosphorus fractions in Karri (*Eucalyptus diversicolor* F. Muell.) forest soils. *Biol. Fertil. Soils* **1992**, *14*, 200–204.

27. Tabatabai, M.A. Soil Enzymes. In *Methods of Soil Analysis*: *Microbiological and Biochemical Properties*; Weaver, R.W., Ed.; SSSA Book Series. No.5; Soil Science Society of America (SSSA): Madison, WI, USA,1994; pp. 775–833.

28. Eivazi, F.; Tabatabai, M.A. Glucosidase and galactosidases in soils. *Soil Biol. Biochem.* **1988**, *20*, 601–606.

29. *SAS/STAT User's Guide*, Version 9.1; Statistical Analysis System (SAS) Institute: Cary, NC, USA, 2005.

30. Zhao, Y.; Wang, P.; Li, J.; Chen, Y.; Ying, X.; Liu, S. The effects of two organic manures on soil properties and crops on a temperate calcareous soil under a wheat-maize cropping system. *Eur. J. Agron.* **2009**, *31*, 36–42.

31. Green, V.S.; Scott, D.E.; Cruz, J.C.; Curi, N. Tillage impacts on soil biological activity and aggregation in a Brazilian Cerrado Oxisol. *Soil Tillage Res*. **2007**, *92*, 114–121.

32. McCallister, D.L.; Bahadir, M.A.; Blumenthal, J.M. Phosphorus partitioning and phosphatase activity in semi-arid region soils under increasing crop growth intensity. *Soil Sci.* **2002**, *167*, 616–624.

33. Juma, N.G.; Tabatabai, M.A. Distribution of phosphomonoesterases in soils. *Soil Sci.* **1978**, *126*, 101–108.

34. Harrison, A.F. Relationship between intensity of phosphatase activity and physico-chemical properties in woodland soils. *Soil Biol. Biochem*. **1983**, *15*, 93–99.

35. Turner, B.L.; Haygarth, P.M. Phosphatase activity in temperate pasture soils: Potential regulation of labile organic phosphorus turnover by phosphodiesterase activity. *Sci. Total Environ*. **2005**, *344*, 27–36.

36. Sun, R.L.; Zhao, B.Q.; Zhu, L.; Xu, J.; Zhang, F.D. Effects of long-term fertilization on soil enzyme activities and its role in adjusting-controlling soil fertility. *Plant Nutr. Fertil.* **2003**, *9*, 406–410.

37. Eivazi, F.; Tabatabai, M.A. Factors affecting glucosidase and galactosidase activities in soils. *Soil Biol. Biochem*. **1990**, *22*, 891–897.

38. Benitez, E.; Nogales, R.; Campos, M.; Ruano, F. Biochemical variability of olive-orchard soils under different management systems. *Appl. Soil Ecol*. **2005**, *32*, 221–231.

39. Biederbeck, V.O.; Campbell, C.A.; Rassiah, V.; Zentner, R.P.; Guang, W. Soil quality attributes as influenced by annual legumes used as green manure. *Soil Biol. Biochem*. **1998**, *30*, 1177–1185.

40. Acosta-Martinez, V.; Harmel, R.D. Soil microbi Soil Science Society of America al communities and enzyme activities under various poultry litter application rates. *J. Environ. Qual*. **2006**, *35*, 1309–1318.