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ORIGINAL RESEARCH

Assessing microbial population dynamics, enzyme activities and phosphorus availability indices during phospho-compost production

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Abstract

Purpose This study assessed changes in bio-quality indices and plant available P released during aerobic–thermophilic co-composting of different mix ratios of non-reactive ground phosphate rock (GPR) with poultry and cattle manures.

Methods Aerobic–thermophilic co-composting of different mix ratios (5:5, 8:2, 7:3 and 9:1) of non-reactive GPR with poultry and cattle manures was carried out. Compost piles without GPR addition were included as control. Compost samples were taken at mesophilic, thermophilic, cooling–stabilization and maturing phases for microbial counts, enzyme activities and P assessment.

Results Abundance of different microbial groups across the composting phases varied greatly ($p < 0.001$) mostly dominated by fungi that was generally more in the cattle than poultry manure-based phospho-composts. Fungi and actinomycetes counts in the composts were positively correlated with alkaline phosphatase and β-glucosidase. A strong inter-correlation between β-glucosidase and alkaline phosphatase ($r = 1.000$, $p < 0.001$) was observed, suggesting that both enzymes possess same origin. Alkaline phosphatase and β-glucosidase contents in the phospho-composts showed negative correlation with water soluble P ($r = -0.65$, $p < 0.001$), and Bray P1 and Fe–P contents ($r = -0.15$, $p > 0.05$) indicating inhibition of the P forms. Quantitatively higher P was obtained from poultry manure-based phospho-compost and in the 8:2 mix ratio at compost maturity. Microbial diversity and enzyme activity exerted positive impact on P mineralization and availability from the non-reactive GPR signifying the beneficial effect of co-composting.

Conclusions Co-composting of P-rich non-reactive GPR with organic wastes containing variable chemical composition promotes microbial diversity during composting and increases plant available P content and compost fertilizer value.

Keywords Phospho-compost · Compost bio-quality indices · Enzyme activities · Available P · Non-reactive phosphate rock

Introduction

With urbanization and rapid increase in human population which was recently projected to hit 11.2 billion in 2100 (UN-DESA 2015), greater improvements in waste management strategies are needed to deal with the rising volume of global wastes generation. Global average daily estimate of 3.5 MT wastes generation is further expected to rise to over 6 MT per day in 2025 (Hoornweg et al. 2013). Available statistics suggest that the amount of solid waste generation and its composition vary greatly across countries; with greater amount in highly developed and wealthy countries including those with high population density (UNEP 2011). Composting is one of the crucial management strategies often used for the safe disposal of the prodigious amount of wastes that are continuously generated globally through anthropogenic

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activities. However, the importance of composting as a waste management option encompasses multiple benefits such as greening of wastes, environmental friendliness and its potential to guarantee safe and sustainable future (UNEP 2011). Aside providing carbon for soil organisms, it also serves as cost-saving potential bio-fertilizer source and the supply of valuable nutrients for plant use (Chatterjee et al. 2013; Manderson 2014).

The use of composts as plant nutrient-sources or as soil conditioners on farmlands, public and private gardens, parks, highway embankments, landscaping and for environmental rehabilitation (Erickson et al. 2009) represents an age long practice (Tognetti et al. 2011; Pan et al. 2012). Thus, it is regarded as one of the important low cost inputs used for meeting nutrient requirements for plant growth (Zameer et al. 2010). However, the success of any composting process as influenced by the quality and usefulness of the compost thereof depends on such factors as the composition of functional microbial community diversity in the wastes, the quality of the organic materials, the effectiveness of mix of the composting materials and favourable physico-chemical conditions during composting including the provision of adequate aeration (Partanen et al. 2010; Chatterjee et al. 2013). Among the major reported groups of microbiological components in composts are bacteria, fungi and actinomycetes that are able to degrade even the more recalcitrant compounds in wastes (Karnchanawong and Nissaikla 2014). Also, critical plant nutrient transformation processes that take place during composting such as nitrogen (N) and phosphorus (P) are microbially mediated (Bernal et al. 2009; Maeda et al. 2013). These microbial activities are achieved through the action of enzymes that are responsible for the hydrolysis of complex macromolecules present during organic waste metabolism (Delgado et al. 2004). The level and rate of microbial activity are, however, determined by the amount of readily metabolizable substrates present in the compost (Yu et al. 2007) as well as temperature, pH and water content under aerobic conditions (Rebollido et al. 2008).

Although the content of plant available P from composts varies considerably depending on the amount of total P present in feedstock, its release rate characteristics as influenced by compost stability are often very slow and ranges between 3 and 22% of the total P content (Prasad 2009). Literature evidence suggests that the possible P surplus may exist when composts are applied to crops as N source (Mikkelsen and Hartz 2008). However, the widespread P-deficiency problem in the world soils (Vance et al. 2003) due to the relatively small pool of native soil P is exacerbated by crop removal and poor management practices such as over-reliance on the sole use of manures that are often applied at low to sub-optimal levels (Kutu 2012). Thus, it is often required that substantial external P addition must be made to satisfy crop demand and guarantee high yield. Regrettably, the problems of limited access to inorganic P fertilizers due to scarcity and high price (FAO 2005) have reportedly contributed to a significant decline in its use particularly by resource-poor subsistence and medium-scale farmers (Bationo et al. 2006) leading not only to massive yield losses but also huge yield gap and food insecurity. Therefore, it is expedient to urgently identify and proffer viable alternatives to the current costly inorganic P fertilizers used for crop production in the form of a cheaper, locally and readily available P-rich and plant available fertilizer product.

Among the numerous scientifically proven, practically inexpensive, low input technology and alternative P fertilizer sources is the co-composting of reactive and non-reactive ground phosphate rocks (GPR), herein described as "phospho-compost technology". Phospho-composting has been reported to offer environmental advantage of safe disposal of organic wastes (Hellal et al. 2013) and, hence, appears attractive especially where the P-rich rock abounds in large quantities. Co-composting is a microbially mediated process that involves the solubilization of P from both organic and inorganic pools through organic acids release from microbial activities in which hydroxyl and carboxyl groups chelate the cation bound to phosphatic rock and finally gets converted into soluble P form (Zaidi et al. 2009). The process has been reported to stimulate the growth of numerous types of bacteria and fungi at different stages of composting that produce large amounts of organic acids and humic substances (Suárez-Estrella et al. 2008) including the release of extracellular enzymes that promote organic matter degradation (Vishan et al. 2014). Other specific P solubilizing microbes (PSM) and acid-producing micro-organisms (APSM) such as *Pseudomonas* spp., *Bacillus* spp., *Aspergillus* spp. *and Penicillium* spp. are also reported to be present during the composting process (Zaidi et al. 2009). Quantification of these microorganisms and enzyme activities during a co-composting process is thus crucial for assessing useful information regarding nutrients transformation and release (Castaldi et al. 2008).

The measure of the content of enzyme activities has been reported as a useful indicator for determining the functional diversity of microbial communities or mass turnover in composts, assessing compost stability, and also ascertaining compost maturity and quality (Gómez-Brandón et al. 2008; Cardoso et al. 2013). This enzymatic activity, particularly phosphatase, plays a critical role in P cycling, serves as a vital microbial indicator for compost quality assessment (Stott et al. 2010; Lehman et al. 2015), and is often synthesized by both microorganisms and plants (Makoi and Ndakidemi 2008). Although several authors have reported changes in the microbial diversity and activity during organic waste composting including animal manures, studies on the co-composting of organic wastes with P-rich phosphate rocks

are scanty. This study assessed changes in bio-quality indices and plant available P released during aerobic–thermophilic co-composting of different mix ratios of non-reactive GPR with poultry and cattle manures.

Materials and methods

Phospho-compost preparation and sampling for laboratory analysis

The preparation of the phospho-composts was done at the University of Limpopo Experimental farm, Syferkuil (23°50′36.86″S and 29°40′54.99″E) using aerobic–thermophilic composting technique (Ahmad et al. 2007). Poultry and cattle manure from a layer pen and beef cattle kraal, respectively, were collected from a nearby private commercial farm at Solomondale, which is approximately 20 km away from the University Campus. The GPR used was obtained from the Foskor mining company, Phalaborwa. Compost piles of the different mix ratios (5:5, 7:3, 8:2, 9:1, w/w) comprising 500 kg (dry basis) were separately prepared on a concrete floor. A total of ten phospho-compost piles were produced including manure heaps with no GPR addition as control. Compost heaps were regularly turned at 2-weekly interval to provide satisfactory aeration with moderate water addition until the end of the thermophilic phase to allow for re-heating of compost heap through substrates degradation by microorganisms. The moisture content of the composts was maintained beneath the water-holding capacity during the composting period so as to prevent inhibition of or slow down microbial activities. Composting was done for a period of 4 months to achieve proper curing. Temperature readings from the different phospho-composts were taken at four different points per heap and recorded prior to compost turning using Hanna instrument (HI 9043) thermocouple thermometer. Quantification of the amount of available P mineralized and the assessment of bio-quality parameters of the composts were done through laboratory analyses and microbial assays, respectively, using compost samples taken at mesophilic, thermophilic, cooling–stabilization and maturing phases. These phases were translated to 4, 8, 12 and 16 weeks following the commencement of the co-composting process. Phospho-compost samples taken were passed through 2 mm sieve and kept at 4 °C until ready for use for the microbial analyses, while sample for P study was air dried, sieved and subsequently used for the determination.

Microbial population enumeration

Population counts for bacteria, actinomycetes and fungi in the various compost samples were done through serial dilution procedure described by Benson (2002) using different sterilized growth media. General heterotrophic plate counts were done on nutrient agar (NA) (Biolab Midrand, South Africa). Actinomycetes were isolated and enumerated on actinomycete isolation agar (Sigma–Aldrich, South Africa). Filamentous fungal counts on malt extract agar (MEA) (Biolab Midrand, South Africa) were used and supplemented with 0.3 mg/l chloramphenicol and 0.5 mg/l streptomycin prepared according to the manufacturer's recommendations. A dilution series that ranged from 10^{-1} to 10^{-5} was prepared in triplicate using 1 g of compost in 9 ml of saline solution and a 100 µL aliquot of each dilution was spread on the isolation plates. The various isolation plates were incubated at room temperature and enumerated after 2 days for bacterial count, and 5 days for actinomycetes and fungal counts expressed as colony forming unit per gram (CFU/g).

Enzyme activity determination

Extracellular enzyme activities of alkaline phosphatase, β-glucosidase and dehydrogenase in compost samples and the control were determined colorimetrically using enzyme-specific procedures (Jackson et al. 2013). Although dehydrogenase analysis was the only assay that is mandatory to be carried out using moist samples, all the three enzymes assays were done on moist samples for the purpose of standardization of results (Tabatabai and Dick 2002). Thereafter, data generated on wet basis were corrected for moisture content for final calculation. The content of β-glucosidase was determined by adapting the procedure described by Acosta-Martinez and Tabatabai (2011). One gram of moist compost sample was incubated at 37 °C for 1 hour with toluene, modified universal buffer pH 6.0, and *p*-nitrophenol-β-D-glucosidase (ρNG), and then shaken for 1 hour with calcium chloride and tris-(hydroxymethyl) aminomethane (THAM) before filtering through a Whatman No. 2v filter paper. The absorbance of released *p*-nitrophenol (ρNP) was tested on a microplate reader at 405 nm immediately after yellow color developed. The dehydrogenase assay was done on all compost samples including control following the procedure described by Von Mersi and Schinner (1991). This assay was conducted using 1 g sieved compost mixed with THAM and iodonitrotetrazolium violet-formazan (INT) (Sigma–Aldrich, South Africa) solution and incubated at 40 °C in the dark for 2 h. Thereafter, the absorbance of the reaction product in INT was read in a glass cuvette on a spectrophotometer at 464 nm after 30 min. The methodology for alkaline phosphatase assay was adapted from Deng and Popova (2011) in which 1 g of moist sieved compost sample was incubated at 37 °C for 1 h with a mixture of toluene, modified universal buffer (MUB, pH 6.5 for acid phosphatase), and ρ-nitrophenol (ρNP). Thereafter, 1 ml

calcium chloride and 4 ml sodium hydroxide were added and the mixture immediately filtered through Whatman No. 2v filters. The absorbance of ρNP was measured immediately after yellow colour development with a microplate reader at 405 nm. All measured values of the enzyme activities were reported in gramme per kilogramme dry weight of compost per hour (g dwt/kg/hr).

Extraction and determination of P forms in compost samples

The quantification of P in each compost sample was done by measuring the various P forms, namely Bray P, water extractable, organic and inorganic P fractions consisting of calcium bound-P (Ca-P) and Iron bound-P (Fe–P), which is also described as iron oxide-impregnated P (P_i) as used in this paper. Bray P1 was determined colorimetric following the method described by Bray and Kurtz (1945); organic P concentration was determined following the modified method described by Wang et al. (2013) while the water extractable P (WSP) content was according to Pierzynski (2000). Extraction and determination of Ca bound-P followed the modified method described by Sharpley (2000), while the iron oxide-impregnated P was determined following the adapted method described by Chardon (2000). All P determinations were carried out in triplicates.

Statistical analysis

Data were analysed using descriptive statistics and analysis of variance (ANOVA) of Statistix 10.0 software. Comparison of different means was done with LSD test ($\alpha < 0.05$). The relationship between the measured P and bio-quality parameters was determined by Pearson correlation analysis.

Results and discussion

Diversity of microbes and enzyme activities across the different composting phases

Table 1 shows that the distribution and diversities of these microbes and enzymes varied significantly across the different composting phases. Bacterial count was highest at the maturing phase while fungi and actinomycete counts were highest during the mesophilic and cooling phase,

Table 1 Microbial counts and the concentration of measured enzyme activities in the phospho-compost samples

Treatments/factors	Microbe population (CFU g^{-1} soil)			Enzyme activity (g dwt kg^{-1} h^{-1})		
	Bacteria	Fungi	Actinomycete	β-glucosidase	Alkaline-phosphatase	Dehydrogenase
Composting phases (A)						
Mesophilic	2.25b	6.43a	5.12d	116.6b	2561ab	426c
Thermophilic	1.44c	5.97c	5.40c	107.6b	2363b	593b
Cooling–stabilization	2.21b	6.07b	6.00a	124.6a	2736a	618a
Maturing	2.73a	5.99c	5.78b	119.4ab	2621ab	618a
LSD ($p = 0.05$)	0.07	0.07	0.06	14.4	315	11
SEM	0.27	0.11	0.20	3.6	78.2	46.3
Manure–GPR mix ratios (B)						
10:0	1.92b	6.11a	5.62a	135.8a	2981a	547a
9:1	1.93b	6.15a	5.60a	125.0ab	2744ab	572a
8:2	2.08ab	6.03a	5.48a	127.2ab	2794ab	572a
7:3	2.15ab	5.96a	5.58a	111.7ab	2452ab	570a
5:5	2.70a	6.33a	5.61a	85.6b	1879b	558a
LSD ($p = 0.05$)	0.74	0.52	0.68	47.64	1046	66
SEM	0.14	0.06	0.03	8.8	192.5	4.9
Manure sources						
Poultry manure	1.72b	5.51b	4.70b	61.4b	1348b	571a
Cattle manure	2.59a	6.72a	6.44a	172.7a	3793a	556a
A × B (F value)	6.18**	2.23*	1.16^{ns}	0.60^{ns}	0.60^{ns}	17.68***

Different letters for mean within column under treatment factors indicate significant difference at $p < 0.05$

LSD = Least significant difference at $\alpha < 0.05$, **significant at $p < 0.001$, ***significant at $p < 0.0001$, ns = not significant ($p \geq 0.05$), PM- and CM-based implies poultry manure and cattle manure-based phospho-compost

respectively. Bacterial counts during the mesophilic and cooling–stabilization phases were significantly comparable. Earlier studies (Yu et al. 2007; Partanen et al. 2010) revealed that microbial number and the level of activity changed during composting leading to heat generation that exert adverse effects on the diversity and distribution of mesophiles. Although few bacteria are reported to be thermo-tolerant, most were more active under cool environment (Devi et al. 2012). The least bacteria and fungi counts were recorded during thermophilic phase possibly due to dormancy and differential responses to compost temperature (Huhe et al. 2017). On the other hand, actinomycete count was lowest during the mesophilic phase which may be attributed to available species during this phase; with some species appearing at the thermophilic phase while others become an important part of the cooling phase (Vishan et al. 2014). Notwithstanding, fungi represented the dominant count across the four composting phases followed by actinomycete. Bacterial count increased by more than 50% after the thermophilic phase and peaked at the maturing phase suggesting possible re-colonization of beneficial thermo-tolerant microbes after the thermophilic phase (Scheuerell and Mahaffee 2005; Villar et al. 2016). This may have been facilitated by aeration and regular turning and watering during the composting process. Similarly, high fungi distribution observed at the mesophilic phase may possibly be attributed to the relatively high moisture content around the compost since fungi as saprophytes form more colonies in moist environment than dry environment (Srivastava et al. 2011). Actinomycete count recorded during the cooling–stabilization phase was highest possibly due to its primary metabolic role in oxidation of recalcitrant materials such as lignin and cellulose during agricultural waste composting (Yu et al. 2007; Partanen et al. 2010; Devi et al. 2012; Limaye et al. 2017). Hence, the diversity of microbial community in compost underpins the role of microbes in the monitoring of co-composting process for nutrient-rich and high-quality product. This assertion is in agreement with earlier reported findings (Bonilla et al. 2012; Villar et al. 2016). Earlier studies (Seshachala and Tallapragada 2012; Zhang et al. 2014) have shown that the population richness and diversity of P-solubilizing bacteria were more in organic matter-rich environments, which ultimately favor both microbial growth and the promotion of microbial P solubilization.

The alkaline phosphatase represented the most dominant enzyme across the different phases, while the activity of β-glucosidase was least (Table 1). Similarly, the activities of alkaline phosphatase and β-glucosidase were more prevalent in the cattle than poultry manure-based phospho-composts while the measured dehydrogenase activities in both phospho-compost types are statistically comparable, albeit marginally (2.7%) higher in poultry manure-based phospho-composts (Table 1). The activity of enzymes in composts provides an indication of the presence of easily degradable organic compounds such as simple sugars (Pane et al. 2011; Bonilla et al. 2012). Hence, a rapid carbon turnover during composting is a function of the activity of β-glucosidase that degrades cellulose with glucose utilized as a source of energy by the inherent microbes (Albiach et al. 2000). Marinari et al. (2000) reported that alkaline phosphatase activity is stimulated by the presence of organic compound and, hence, the dominance of phosphatase activity in the phospho-compost ratios. Previous studies (Srivastava et al. 2012; Uz and Tavali 2014) have also suggested that organic matter increases soil β-glucosidase and alkaline phosphatase activity depending on the composition of carbon compounds. Although β-glucosidase activity was highest during the cooling phase, measured concentrations in compost samples at the cooling and maturing phases were statistically comparable and marginally (4.4%) lower at compost maturing phase (Table 1). The activities of alkaline phosphatase and dehydrogenase measured were highest at the cooling phase but statistically comparable to the measured value at both mesophilic and maturing phases. Dehydrogenase was the least active at the mesophilic phase of co-composting process.

Microbial counts and enzyme activities in the different phospho-compost types

The influence of manure types and its various mixed ratios with non-reactive GPR on microbial counts and the content of enzyme activities are presented in Table 1. The manure and GPR mix ratio of 5:5 recorded the highest bacteria count, which differed significantly from the count obtained in compost heap with no GPR addition. The implication is that the mix ratio enhanced the proliferation and activity of P solubilizing microbes through the synergy between the activities of P solubilizing microbes and the organic materials that are present in the compost heaps. Regrettably, the highest microbial count measured in the 5:5 phospho-compost mix ratio did not translate to the highest plant available P content. Zhang et al. (2014) reported that adding small amounts of inorganic phosphorus to the rhizosphere could drive phytic acid mineralization by bacteria. Such microbes are able to solubilize P from the non-reactive GPR through the production of protons and organic acid, thereby creating acidity that promote the mineralogical dissolution and chelating of calcium and P contained in the ground rock (Chien et al. 2010; Edwards et al. 2010). The different manures and GPR mix ratios exerted significant effects on bacterial count. While the variation in manure type and GPR mix ratios had inconsequential effects on bacterial count, the measured fungal and actinomycete counts were quantitatively highest in the 5:5 and 10:0 mix ratios, respectively.

Albeit, microbial counts were generally higher in the cattle than poultry manure-based phospho-composts suggesting greater nutritional conditions in former system to sustain growth and overcome the screening effect of high temperature (Huhe et al. 2017).

The different manures and GPR mix ratios exerted significant effects on the content of β-glucosidase and alkaline phosphatase activities. The activities of β-glucosidase and alkaline phosphatase were highest in compost without GPR addition although the values were statistically comparable to the measured activities in all other mix. Increasing the amount of GPR addition in the phospho-compost heaps resulted in an increased bacterial count while such increases resulted in decreased contents of β-glucosidase and alkaline phosphatase activities. Overall, the mean microbial count and enzyme activities in the various mix ratios were generally higher in the cattle manure-based than poultry manure-based phospho-composts. The greater increase in enzyme activities in the phospho-composts with higher organic manure content suggests better condition and increased content of degradable compounds for the extracellular enzyme activities. Similar observation by Chang et al. (2007) showed that the enzymatic activities are significant and linearly correlated with soil organic matter contents. The implication is that increasing the proportion of manure in the mix ratio relative to the GRP content would increase enzymatic activities through the formation of freer enzyme complexes. The different manure and GPR mix ratios had no significant effect on the dehydrogenase activity. The measured content of dehydrogenase activity reflects the total range of oxidative activity of the microflora and, hence, used as an indicator of microbial activity. This probably explains the distribution of fungi and actinomycetes in the phospho-compost that were appreciably not strongly influenced by added GRP as alluded to by Marinari et al. (2000).

Phosphorus concentrations measured in the different phospho-composts

The concentration of the various P forms and inorganic P fractions measured differed significantly across the different phases of phospho-compost production (Table 2). Organic P concentration was highest at the mesophilic phase and decreased steadily during the co-composting process until the cooling–stabilization phase, after which, the content increased by over 49% at compost maturing. However, the concentration of WSP, Ca-bound P and Fe–P fractions in the compost samples increased steadily as the co-composting process progressed, and peaked in the mature or cured product. The concentration of Ca bound-P at the compost maturing phase was nearly 8 times higher than the concentration at mesophilic phase. On the other hand, the concentrations

Table 2 Phosphorus concentrations (mg kg^{-1}) measured at each composting phase during phospho-compost production and and its mixed ratios

Treatments/factors	Bray P1	Organic P	Ca-P	WSP	P_i
Composting phases (A)					
Mesophilic	7.58d	20.4a	108d	989d	393d
Thermophilic	109.6c	15.2c	251c	1577c	2619c
Cooling–stabilization	154.5a	11.0d	711b	1743b	4206b
Maturing	151.7b	16.4b	826a	1870a	4432a
LSD ($p=0.05$)	1.19	0.7	25	16	153
SEM	34.32	1.93	174.1	195	931.7
Manure–GPR mix ratios (B)					
10:0	34d	3.1d	36c	117b	1817b
9:1	86c	12.4c	293bc	1643ab	2845b
8:2	151a	20.6ab	971a	1783a	5039a
7:3	148ab	18.2b	504b	1655ab	2919b
5:5	111bc	24.5a	293ba	1469ab	1944b
SEM	22	3.7	156	308	577
LSD ($p=0.05$)	39.46	4.3	282	498	1349
Manure source					
PM-based	111.5a	19.0a	683a	2287a	3802a
CM-based	100.2b	12.5b	265b	803b	2023b
A × B (F value)	87.7***	9.39***	15.11***	1.42^{ns}	11.83***

Different letters for mean within column under treatment factors indicate significant difference at $p<0.05$

LSD = least significant difference at $\alpha<0.05$, WSP = water soluble P, P_i = iron impregnated P (Fe–P), Ca-P = Calcium bound P. **significant at $p<0.001$, ***significant at $p<0.0001$, ns = not significant ($p\geq0.05$); PM- and CM-based implies poultry manure and cattle manure-based phospho-compost

of WSP and Pi at compost maturing were nearly double and over 11 times higher, respectively, at maturing relative to the concentration measured at the mesophilic phase. The concentration of available P measured increased sharply after the mesophilic phases until the cooling phase but decreased marginally (1.85%) at compost maturity. Overall, the measured concentration of the different P forms was generally lowest at the mesophilic phase while the measured organic P concentration at the different composting phases was lower than any other P forms. Similarly, the measured P forms and fractions were generally higher ($p \leq 0.05$) in the poultry manure than in the cattle manure-based phospho-composts (Table 1).

Interaction effect of composting phases and manure–GPR mix ratios on compost quality indices

The combined effect of compost sampling phases and manure–GPR mix ratios on bacterial and fungi counts and dehydrogenase activity was significant ($p < 0.001$) but had inconsequential effect on the actinomycete count and alkaline phosphatase and β-glucosidase activities (Table 1). The effect on the concentration of P forms was similarly significant except for the WSP fraction (Table 2). Bacterial and fungal counts were, therefore, highest in manure–GPR mix ratio of 5:5 during the mesophilic phase while actinomycete count was highest in compost-GPR mix ratio of 8:2 at the cooling–stabilization phase (Fig. 1). The activity of dehydrogenase was comparable across the different mix ratios and composting phases but marginally highest in the 9:1 compost-GPR mix ratio at the thermophilic composting phase (Fig. 2). However, the activities of both the alkaline phosphatase and β-glucosidase were statistically highest in the compost-GPR mix ratio of 8:2 at the cooling–stabilization phase. Available P1 concentration measured was highest in compost mix ratio 7:3 at the cooling–stabilization phase but was significantly comparable with the measured concentration in compost mix ratio of 8:2 beyond the thermophilic phase (Fig. 3). The concentrations of WSP, Ca-P and Fe–P were highest in the 8:2 compost-GPR mix ratio at the maturing phase except with Fe–P that was highest at the cooling phase. The organic P concentration from the compost-GPR mix ratio of 5:5 at the mesophilic phase was 17.6% more than that obtained in the compost-GPR mix ratio of 8:2 at the same mesophilic composting phase (Fig. 3).

Pearson correlation matrix among measured compost bio-quality parameters and P forms

Results of the Pearson correlation matrix among various bio-quality parameters and the concentration of P forms and fractions in the compost sampled during composting are

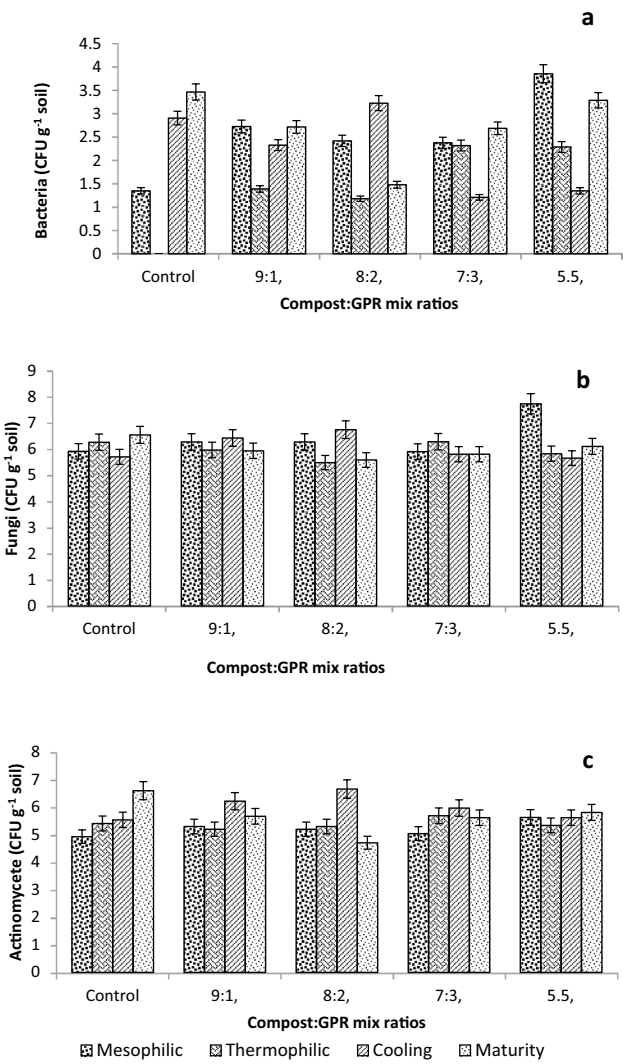

Fig. 1 Phospho-compost × composting phase interaction effects on microbial count (where **a** = bacteria; **b** = fungi; **c** = actinomycetes)

shown in Table 3. Bacterial count in the different phospho-composts showed fairly good but significant correlation with fungi and actinomycete ($r \leq 0.52$; $p < 0.001$) but poor correlation ($r = 0.27^{**}$) with the evaluated alkaline phosphatase and β-glucosidase as extracellular enzyme activities. Both fungi and actinomycete colony count showed strongly significant correlation with each of alkaline phosphatase and β-glucosidase activities ($r > 0.70$; $p < 0.001$). The relationship between alkaline phosphatase and β-glucosidase was very strong and significant ($r = 1.000$, $p < 0.001$), suggesting that both are from the same origin. Fungi population count showed poorly negative correlation with dehydrogenase activity and Fe–P content ($r \leq 0.28$; $p < 0.05$) but strongly negative correlation with and Bray P1 content ($r = -0.61$; $p < 0.001$). The activities of alkaline phosphatase and β-glucosidase were significant and negatively correlated to

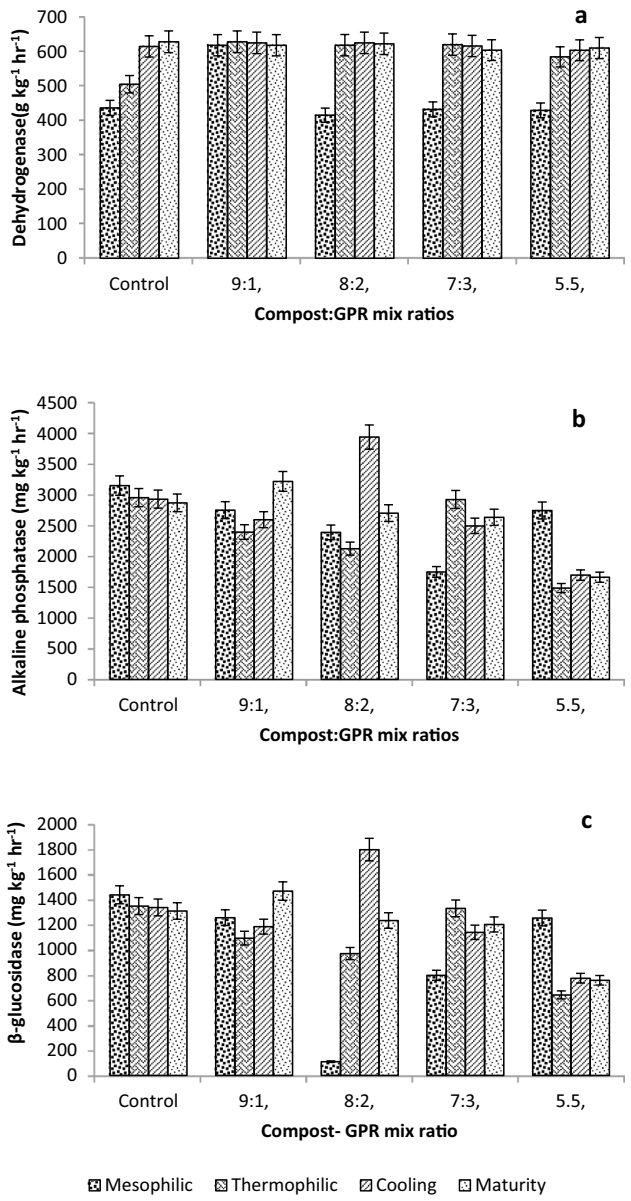

Fig. 2 Phospho-compost × composting phase interaction effects on the activities of the measured enzymes (where: **a** = dehydrogenase; **b** = alkaline-phosphatase; **c** = β-glucosidase)

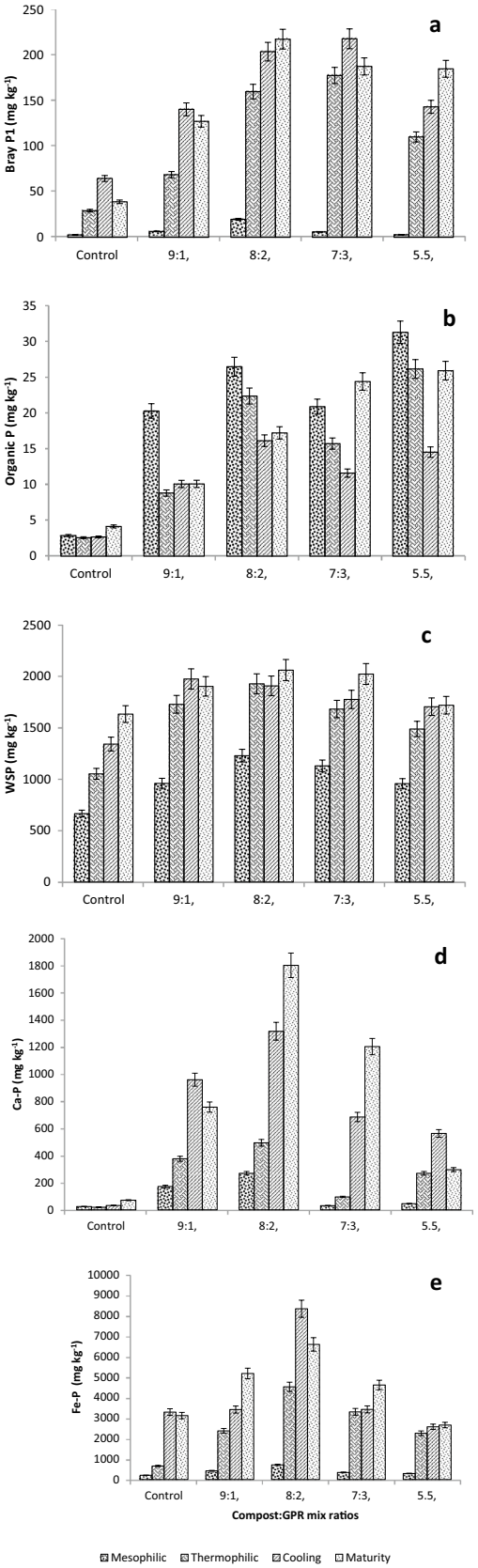

Fig. 3 Phospho-compost × composting phase interaction effects on the various P forms, namely **a** Bray P1, **b** organic P, **c** water soluble P; and inorganic P fractions consisting of **d** Ca-P, and **e** Fe–P

the water soluble P concentration while Fe bound-P showed strongly significant correlation with WSP and Bray P as well as the activities of dehydrogenase enzyme.

Conclusion

Microbial counts and enzyme activities are key bio-quality parameters used for monitoring compost maturity. The diversity and distribution of microbial community at the identified composting phases vary greatly with generally lower bacterial counts recorded during the thermophilic phase than either of

Table 3 Pearson correlation matrix between microbial counts, activities of enzymes and P availability indices of the different phospho-composts

Parameters	Bacteria	Fungi	Actino	A-phosp	β-glucos	Dehyd	Bray P1	WSP	P_i
Bacteria	1								
Fungi	0.52***	1							
Actino	0.49***	0.77***	1						
A-Phosp	0.27**	0.70***	0.73***	1					
β-glucos	0.27**	0.70***	0.73***	1.00***	1				
Dehydr	0.08	−0.28**	0.12***	−0.08	−0.08	1			
Bray P1	0.00	−0.23*	0.11	−0.06	−0.06	0.70***	1		
WSP	−0.25**	−0.61***	−0.49***	−0.65***	−0.65***	0.44***	0.48***	1	
p_i value	0.08	−0.25***	0.09	−0.16	−0.16	0.57***	0.66**	0.61***	1

*, ** and *** indicate significant at 5, 1 and 0.1% probability level, respectively

Actino = actinomycete, A-phosp = alkaline-phosphatase, β-glucos = β-glucosidase, Dehyd = dehydrogenase, WSP = water soluble P, P_i = iron impregnated P (Fe–P)

fungi or actinomycete counts. The action of these microbes exerts remarkable impact on compost quality and its ultimate usefulness in agriculture. Amending organic wastes with P-rich non-reactive GPR during co-composting exert influence on microbial counts and the activities of β-glucosidase and alkaline phosphatase in the mature/cured compost, and also resulted in elevated plant available P content and fertilizer value of the compost product. The study provides novel findings with diverse implications. It constitutes both relief and benefits for millions of resource-poor farmers who regularly experience restricted access to readily available and affordable P-rich fertilizers, and burdened by over-reliance on expensive and environmentally unfriendly synthetic P fertilizers to address land degradation challenges on their fields. These often result in reduced productivity and massive crops yield gap. For the phosphate rock mining industries, it could promote or stimulate new local markets for the utilization of unprocessed raw ground rock in the compost industries, while the latter would also benefit through access to cheaper raw material (GPR) for improving the P content of composts. Ongoing research works include the quantification of plant available P from phospho-compost produced under field conditions and the assessment of the risks of trace metal load in soil and crops following continuous soil amendment with phospho-composts produced from non-reactive GPR.

Compliance with ethical standards

Conflict of interest The authors declare that there is no conflict of interest.

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