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ACKNOWLEDGEMENTS

Foremost, I would like to express my deep sense of gratitude and indebtedness to my principal supervisor, Prof. Petra Marschner for her expert guidance, patience, motivation, valuable suggestions, constructive criticism and amicable behaviour throughout my PhD journey which led this work to a successful completion. I cannot imagine having a better supervisor for my PhD. My sincere gratitude is also to my cosupervisor, Chris Penfold (The University of Adelaide) for his constructive comments, criticism and encouragement to complete this research.

I also express my sincere thanks to Mr Colin Rivers for his help in soil collection and technical matters.

Special thanks to SARDI Greenhouse officers, Paul Ingram and Andreas Flenche for arranging the greenhouse facilities.

Thanks to former and current members of Soil Organic Matter group (Raj, Tra, Mavi, Nasrin, Hasnuri, Hasbullah, Alamgir, Andong, Nan, Malik and Ta) for their support and encouragement throughout my study. Thanks also to the Soils group, School of Agriculture Food and Wine for valuable friendship and networking.

I would like to thank my family members for their support and encouragement. Last but not least, my sincere thanks to my better half, Prem Kumar for his support and unconditional love throughout the years. Thank you with all my heart.

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ABSTRACT

Deficiency of plant available phosphorus (P) is common in many soils, therefore application of inorganic P fertilisers is used to overcome P deficiency. The low use efficiency of inorganic P fertilisers has increased interest in the use of alternative sources of P. Application of organic materials like manure, plant residues and compost has been shown to increase P availability and inorganic fertiliser use efficiency and diverts these wastes from landfill. Little information is available about the effect of composts prepared from different feed stocks and with different particle sizes on P availability and P uptake by plants in different soils or their effect on rock phosphate solubility in neutral and alkaline soils. The aims of this study were to (i) determine the effect of different composts on soil P pools and P uptake by wheat, (ii) compare the effect of composts types and application methods on solubility of rock phosphate and P uptake by wheat, (iii) determine the effect of compost particle size on P pools and microbial biomass in soils differing in pH, clay and organic matter content.

An experiment with different composts was conducted in a loamy sand soil with low P availability and wheat was grown until grain filling. The composts were derived from animal manure and straw (C1), garden waste (C2), wood chips and bark (C3) or kitchen waste (C4) and differed in total C, N and P as well as in available P concentration. Soil P pools, soil respiration, plant dry weight and P uptake were measured on days 14, 28 and 72. Composts with high P availability increased labile P pools more than those with low P availability. From 14 days onwards, P availability decreased and organic P and less labile P pools were formed. Wheat P uptake was highest with fine textured composts with low C/N and C/P ratio and lowest with coarse textured compost.

Two experiments were conducted using different composts applied as mulch or incorporated into the loamy sand soil into which rock phosphate was mixed and wheat was grown until grain filling. The composts C1 (from garden waste) and C2 (from wood chips and bark) differed in C/P ratio, C/N ratio, total P and available P concentrations. Compared to rock P alone, the combination of compost and rock P increased the concentration of available P (microbial P and NaHCO₃-Pi) and plant P uptake. However when compost was incorporated in soil with rock phosphate, the concentration of available P and plant P uptake were higher than expected by the addition of P with the compost alone. Thus, compost incorporation increased the concentration of compost and rock phosphate also increased the concentration of NaOH-Pi and HCl pools indicating effects on P flux between pools. These compost effects were greater when the compost was incorporated compared to application as mulch.

An incubation study was carried out to compare the effect of compost particle size on soil P pools and microbial biomass in soils differing in pH, clay and organic matter content. Compost from garden waste was sieved to particle sizes > 5 mm, 3-5 mm and < 3 mm and these fractions were mixed into three different soils at the same rate of total P added (50 mg P kg⁻¹ soil). Soil respiration and microbial biomass and soil P pools were measured on days 25 and 50. Soil respiration was higher with the coarser particles whereas microbial biomass was greater with finer fractions. The size of the P pools was mainly affected by soil type. The effect of the compost was small but the compost fractions differed in their effect on available P with a greater increase by the finest fraction compared to the coarser fraction. Irrespective of compost addition, the acidic soil had the highest concentration of Al and Fe associated P, whereas Ca associated P dominated in the alkaline soil. The size of most P pools including so-called stable P

pools like NaOH-P and HCl-P changed over time in amended and non-amended soils. It can be concluded that the size of P pools is more strongly affected by soil properties such as organic matter content, texture and pH than by the compost fractions.

A glasshouse experiment with four soils (K1, K2, B1 and B2) with different properties was conducted to compare P uptake of wheat and the size of soil P pools in four soils differing in pH, organic matter content and texture. Wheat was grown for 63 days with or without compost (from garden waste) applied as layer on the soil surface. The treatments also included unamended soils. Plant P uptake was greatest in K2 and B2 soils with high organic matter content and higher in plants amended with compost compared to the plants without compost. The concentration of most P pools was higher in the heavier textured soils (K2 and B2). On day 63, the presence of plants increased the concentration of water-soluble P in soils K2, B1 and B2 but only when not amended with compost. The concentrations of microbial P and NaHCO₃-Pi were higher in the planted than the unplanted soil irrespective of compost addition. On the other hand, the NaHCO₃-Po concentration in soils K1, K2 and B2 was lower in the planted compared to the unplanted soil. Thus, although the plants removed P from the soil via P uptake, there was no consistent decrease in P pool concentrations and the concentrations of some of the inorganic P pools (NaOH-Pi, residual P) and NaOH-Po were greater than in the unplanted soil on days 0 and 63. This suggests that plants stimulate the formation of stable inorganic and organic P pools possibly by increasing microbial activity. Compost addition to the plants increased plant growth and P uptake but had little effect on the P pool concentration even in soil B1 which had the lowest P pool concentrations. This suggests that the P added with the compost was readily taken up by the plants and was not converted into soil P pools by the end of the experiment.

DECLARATION

This work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due references has been made in the text. I give consent to this copy of my thesis when deposited in the University Library, being available for loan and photocopying, subject to the provisions of the Copyright Act 1968. The author acknowledges that copyright of published works contained within this thesis (as listed below) resides with copyright holder (s) of those works. I also give permission for the digital version of my thesis to be made available on the web, via the University's digital research repository, the Library catalogue, the Australasian Digital thesis Program (ADTP) and also through web search engines, unless permission has been granted by the University to restrict access for a period of time.

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Date: 05/07/2013

Chapter 1

Introduction and Review of Literature

1.1. Introduction

Phosphorus (P) is the most important nutrients after nitrogen for plant growth and metabolism. Phosphorus plays key roles in plant metabolism and growth such as energy transfer and storage, respiration, photosynthesis and enzyme regulation and as structural component of nucleic acids (DNA and RNA) and membranes (Schachtman et al. 1998; Raghothama 1999).

In nature P exists in living organisms, water, soil and minerals and is widely distributed, but not found in elemental form. Phosphorus is taken up by plants from soils and incorporated into organic compounds (Busman et al. 1997). When plant materials are returned to soils, inorganic P is released by mineralisation of organic P and converted into a number of soil P pools. The soil P concentration is influenced by type of parent material from which soil is derived, degree of weathering and climatic conditions (Fuentes et al. 2008). Although most soils contain high concentrations of total P, very little is available to plants. Phosphorus deficiency is a main constraint to crop production and the second most important problem to soil fertility throughout the world (Vance et al. 2003). The low availability of P in the soil limits plant growth which results in the use of inorganic fertilisers to obtain maximum yield. In the most common inorganic P fertilisers, P is quite soluble and available to plants, but when the fertilisers comes in contact with the soil, various reactions take place and make P less soluble and available (Guo et al. 2000; Ayaga et al. 2006; Vu et al. 2008). Therefore, fertiliser efficiency is very low, in the year of application only about 20% of the applied P is taken up by the crop (Bolland and Gikes 1998; Damodar Reddy et al. 1999). Long-term fertiliser application can result in accumulation of total P which can lead to particulate P in surface runoff and water eutrophication (Correll 1998; Song et al. 2007; Maftoun and Moshiri 2008). The reactions of fertiliser P in the soil are influenced by soil properties like soil pH, temperature, moisture content and types of minerals present in the soil. Rock phosphate, which is the key ingredient for synthesis of inorganic fertiliser, is a limited and non-renewable resource and with current consumption rate these reserves could be exhausted in next 50 to 100 years (Runge-Metzger 1995; Steen 1998). Cost and environmental risk associated with use of inorganic fertilisers have increased the interest in using organic P sources such as manure, plant residues and compost.

Recycling of organic wastes may help maintaining soil fertility and environmental quality as well as reducing the risk associated with dumping of organic waste. Composting is a simple and economic way of reducing organic waste going into landfill (Traore et al. 1999; Speir et al. 2004). When applied to a soil, composts influence soil P availability (Singh and Jones 1976; Kwabiah et al. 2003) but the effect depends on compost properties (type of feedstock, P concentration) as well as on soil properties and climatic conditions (Dorado et al. 2003). Amendment of soil with organic materials like manure, compost and plant residues can increase the soil P availability directly by the P contained in them and indirectly by release and production of organic acids, stimulation of microbial activity or blocking of P fixing sites which can increase P mobilisation and decrease P fixation (Ayaga et al. 2006; Fuentes et al. 2006; Khan and Joergensen 2009). Some limitations of the wide-spread use of compost as alternative nutrient source are the high product and transport costs as well as the variable composition of the composts.

To increase the crop production and reduce cost, inorganic fertiliser can be combined with organic amendments. Rock phosphate is composed of Ca-phosphates and has a low solubility in neutral and alkaline soils (Banger et al. 1989) whereas its solubility is higher in acidic soils (Hongqing et al. 2001). Solubility can be increased by cocomposting rock phosphate with manure or plant residues (Medhi and De Datta 1996; Zaharah and Bah 1997; Tian and Kolawole 2004; Akande et al. 2005). This increase in rock phosphate solubility can be explained by the release of organic acids during decomposition of organic materials (Singh and Amberger 1998; Kolawole and Tian 2010). In this review the effect of compost on P availability of the soil will be outlined.

1.2. Phosphorus

1.2.1. Forms of Phosphorus in soil

Phosphorus in the soil comes from both anthropogenic and pedogenic sources including use of inorganic fertiliser, decomposition of organic materials and weathering of parent material (Scherer and Sharma 2002; Verma et al. 2005). Soil P is classified into two groups: organic (Po) and inorganic P (Pi) and the solubility of most these P forms are very low in soils. Although the total soil P concentration is often high (around 2000-3000 mg P kg⁻¹ soil), the concentration of available P in soil solution is very low (<0.01 to 1 mg L⁻¹ in highly fertile soils) representing a small percentage (<1%) of total soil P (Richardson et al. 2005) and (Jones and Oburger 2011). Only the orthophosphate in the soil solution can be taken up by plants and occurs as dihydrogen phosphate ion (H₂PO₄⁻) in acidic soils and as hydrogen phosphate ion (HPO₄²⁻) in alkaline soils. The release of orthophosphate anions from organic P (Po) and inorganic P (Pi) is influenced by dissolution-precipitation, sorption-desorption, mineralisation-immobilisation and soil pH (Celi et al. 2001; Griffin et al. 2003; Mkhabela and Warman 2005).

Inorganic P in the soil is mainly found associated with Al and Fe oxides, Ca, clay minerals and organic matter (Richardson et al. 1994). The inorganic fraction includes

170 forms ranging from very immobile to labile forms (Holford 1997). In unweathered or moderately weathered soils with neutral or alkaline pH, primary minerals like apatite, hydroxyapatite and francolites are dominant, whereas P associated with Al and Fe oxides predominates in soils with acidic pH, (Sims et al. 2005). Therefore the forms and solubility of inorganic P are strongly influenced by soil properties like soil pH, concentrations of Ca, Al and Fe and soil texture and organic matter content.

Organic P is an important form of P for plant uptake and 20-80% of total P in soil found in organic form (Richardson et al. 1994). Organic P in the soil is derived from microorganisms and organic materials like plant material and animal waste. The main stable organic P compounds are inositol phosphates (\geq 80% of total organic P), phospholipids (0.5-7% of total organic P), nucleic acids (< 3% of total organic P), orthophosphate diesters, monoesters and organic polyphosphates (Turner et al. 2002; Jones and Oburger 2011). Mineralisation by phosphatase released by soil microorganisms and plant roots converts organic P to plant available P (Fuentes et al. 2008) and is influenced by soil pH, moisture, temperature and surface area of soil particles (Shen et al. 2011).

The soil microbial biomass (composed mainly of bacteria and fungi) is also considered as an important source of organic P (Stewart and Tiessen 1987). The soil microbial biomass plays an important role in P dynamics through biochemical transformation of organic matter. Microorganisms mediate several processes in the P cycle and solubilise poorly soluble P forms. The microbial biomass P comprises 0.5 to 26% of total soil P (Oberson and Joner 2005) and is a potentially available P pool.

The release of inorganic P from organically bound P is influenced by interactions among plants, microbes and organic matter in processes such as mineralisation, immobilisation and redistribution (Stewart and Tiessen 1987). Mineralisation refers to the conversion of organic P to inorganic P while immobilisation is the uptake into the microbial biomass which decreases inorganic P concentrations. Mineralisation and immobilization occur simultaneously in the soil. Redistribution occurs when immobilised P is converted into mineralised P and vice versa.

1.2.2. Characterisation of soil P

The correct determination of P in the soil is very important. There are many methods used for determination of soil total P. The most common and generally accepted methods are digestion with perchloric acid (HClO₄), sodium carbonate fusion (Na₂CO₃) (Jackson 1958), sodium hypobromite (NaOBr) fusion followed by H₂SO₄ dissolution (Dick and Tabatabai 1977) or digestion with sulphuric acid-hydrogen peroxide-hydrofluoric acid (H₂SO₄-H₂O₂-HF) (Bowman 1988). Among these methods, perchloric acid (HClO₄) digestion and sodium carbonate fusion (Na₂CO₃) are most common. These methods convert organic P to inorganic P to allow total P determination (Kuo 1996).

There are also various methods to determine plant available P in the soil. In the early 1950's Bray and Kurtz P-1 (Bray and Kurtz 1945) and Mehlich-1 (Mehlich 1953) methods were introduced for estimating available P in acidic and neutral soils. But these methods were unreliable for alkaline or calcareous soils. Later the Mehlich-1 method was modified to Mehlich-3 to cover a wide range of acidic and alkaline soils (Mehlich 1984). In 1954, Olsen et al. (1954) introduced a method which uses 0.5 M NaHCO₃ (at pH 8.5) to extract Ca-bound and adsorbed P in alkaline or calcareous soils. The Olsen method was modified by prolonging shaking time from 30 minutes to 16 hours by Colwell (1963). In Australia, the Colwell method is commonly used for determination

of available P. More recently, the use of anion exchange resin strips for available P determination was introduced by Kouno et al. (1995) and is suitable for various types of soils. The P concentration in the extracts is usually determined by the colorimetric procedure developed by Murphy and Riley (1962).

Organic P compounds are major components of soil P therefore it is important to understand the behaviour of organic P in soils and how this affects on soil P fertility (Dalal 1977). However, it is not possible to measure organic P directly by most methods. Usually, organic P is converted into inorganic P in one soil subsample and then total P is measured. In another subsample total P is measured without conversion of organic P and the difference in total P between these two subsamples is taken as organic P concentration. Extraction of soil with NaOH/EDTA followed by solution ³¹P NMR (nuclear magnetic resonance spectroscopy) can be used to detect the composition of organic P (Vestergren et al. 2012).

Sequential extraction schemes were developed to separate different P fractions based on their chemical solubility. The first method developed by Chang and Jackson (1957) was used for soil inorganic P fractions to detect transformation of applied chemical fertilisers. Bowman and Cole (1978) developed a sequential fractionation method for organic P determination in soils. A comprehensive fractionation scheme was developed by Hedley et al. (1982) to determine soil organic and inorganic P based on their chemical solubility and involved sequential extraction with 0.5M NaHCO₃, 0.1M NaOH, 0.1M NaOH with ultrasonic dispersion, 1.0M HCl and residual P is the P in the soil remaining after the extraction. The fractionation scheme separates soil P into labile, moderately labile and non-labile fractions. For example, sodium bicarbonate P is assumed to be more labile than the sodium hydroxide P. This method was modified by Tiessen and Moir (1993), in which resin P represents the most labile P pool that comes either from soil solution or P adsorbed onto carbonates/hydroxides. Inorganic P (Pi) in NaHCO₃ extract is considered as weakly sorbed P and organic P (Po) in this fraction is easily hydrolysable P. The Pi in the NaOH extract is associated with amorphous Al and Fe phosphates and clay minerals, whereas the Po is associated with fulvic and humic acids (resistant forms). The diluted HCl-P (moderately labile P pool) is associated with Ca, while P in more stable forms is extracted with concentrated HCl. Residual P which is left in the soil after the final extraction step is considered as highly resistant and non-labile (Tiessen and Moir 1993). There is considerable flux among these pools. Available P added to the soil with inorganic fertilisers is initially accumulated as labile and moderately labile P pools and can later be converted into poorly or non-labile P (Vu et al. 2008). On the other hand, residual P is converted into labile to moderately labile P pools (Aulakh and Pasricha 1991). Limitations of the sequential fractionation method are the long time needed for the fractionation, the increasing error with each step and the uncertainty whether the P pools extracted represent P forms present in the soil.

A new method that does not require extraction of P from the soil is X-ray absorption near edge structure (XANES) (Sato et al. 2005; Güngör et al. 2007;). In this technique, synchrotron radiation is applied to excite electrons and these electrons create a spectrum which is detected. The radiation needed for electron excitation is element specific. This method detects P directly in the soil and should therefore better characterise soil P, however it only detects inorganic P forms and is possible only in a synchrotron which is currently available only in a few locations world-wide and its use quite expensive.

1.3. Inorganic and organic P fertilisers

The fate of inorganic fertilisers in soil is dependent on soil properties and environmental conditions (Eghball et al. 1996). With dissolution of inorganic fertiliser (in the presence of water), the P concentration in the soil solution increases. Soluble P ions migrate away from the location where the inorganic fertiliser was placed and are adsorbed to soil particles or combined with Al and Fe in acidic soils and with Ca in alkaline soils to form poorly soluble compounds (Wang et al. 2007; Vu et al. 2008). Thus plants take up < 20% of the applied P in the year of application (Damodar Reddy et al. 1999). As mentioned above, overuse of inorganic fertilisers causes accumulation of stable P forms in the soil and can result in environmental damage like eutrophication (Song et al. 2007; Maftoun and Moshiri 2008). Further, inorganic P fertilisers are synthesised from rock phosphate which is a non-renewable resource.

To increase sustainability of production and maintenance of natural reserves, it is important to use integrated nutrient management systems in agriculture by combining organic and inorganic fertilisers.

The incorporation of organic wastes like manures, plant residues and compost can increase P and N availability, improve soil structure and water holding capacity, soil aggregation and increase microbial biomass. The increase in P availability of soil amended with organic materials depends on their P content, composition and environmental conditions (Bernal et al. 2009). The P concentration in compost can be increased by co-composting of rock phosphate (Zaharah and Bah 1997; Akande et al. 2005). Organic materials can increase soil P availability through various mechanisms: release of soluble P (inorganic P or organic P), change in soil pH (due to the change in ion exchange capacity with increase in organic matter), production and release of organic anions during decomposition, enhanced microbial and enzymatic activities and complexation of Al and Fe ions (Iyamuremye et al. 1996; Ayaga et al. 2006; Fuentes et al. 2008; Khan and Joergensen 2009). Application of organic materials stimulates synthesis of microbial biomass and microbial metabolites. This microbial biomass

requires P for cell components and therefore causes immobilisation (uptake of P by the biomass). This P becomes plant available during biomass turnover.

1.4. Compost and its effect on soil P availability

Direct land application of animal manure and municipal waste to agricultural land is increasing but can be problematic because of pathogen or metal contamination of these materials and high transport costs. To reduce the pathogen risk, improve handling and storage and also to reduce the amount of organic material applied, composting is increasingly employed (Benito et al. 2003; Bernal et al. 2009). Composting transforms organic waste into a more uniform and biologically stable product that can act as slow release source of plant nutrients (Kraus and Warren 2000). Compost is an economic and safe way for treatment of organic waste and has high concentrations of organic matter and available nutrients (Garrido Hoyos et al. 2002; Speir et al. 2004). The increase in P availability with compost application depends on type of feedstock, P concentration, particle size, C/P and C/N ratio and environmental conditions. Combined application of compost with inorganic fertilisers can increase efficiency of inorganic fertilisers and reduce the cost (Sikora and Enkiri 2000; Cornish 2009). To reduce the need for manufacturing soluble P fertilisers, direct use of low grade rock P with organic materials like compost and plant residues may increase plant P availability by increasing the solubility of rock P.

1.5. Aims of this study

Previous studies suggested that organic materials such as manures and plant residues can increase P availability and that combined application of inorganic fertilisers and organic materials may increase efficiency of inorganic fertilisers by influencing soil properties and reducing the formation of stable inorganic P forms (Celik et al. 2004). However little is known about the effect of compost prepared from different feedstock on P pools in different soils and also it is not well understood if compost can improve solubility of rock phosphate when both are applied to soil. Therefore, the objective of this study is to measure changes in soil P pools after compost application.

Specifically the aims are:

- Assess the effect of composts derived from different feed stocks on soil P pools and P uptake by wheat and relate the observed effects to compost properties.
- Compare the effect of different composts on solubility of rock phosphate, soil P pools and P uptake by wheat, when compost is applied as mulch or incorporated
- Determine the effect of compost particle size and soil properties on microbial biomass and activity and P pools.
- Assess the effect of plants and composts on soil P pools in soils with different texture and organic matter content

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Chapter 2

Composts vary in their effect on soil P pools and P uptake by wheat

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Composts vary in their effect on soil P pools and P uptake by wheat

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Performed experiment, analyses of soil, compost and plant samples, data analysis and interpretation, wrote manuscript, acted as a corresponding author.

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Composts vary in their effect on soil P pools and P uptake by wheat

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Abstract

With diminishing world reserves of P deposits and rising fertilizer prices, it is important to find alternative sources of P for crops. Fresh plant residues, manures and composts may supply P to plants, but little is known about the effect of composts derived from different feed stocks on labile and less labile soil P pools. A 72-day glasshouse study was conducted in a loamy sand with low P availability to evaluate the effect of four different composts C1 (from animal manure and straw), C2 (from garden waste), C3 (from wood chips and bark) and C4 (from kitchen waste) on soil P pools and P uptake by wheat 14, 28 and 72 days after compost application. The composts were applied as a 2.5 cm thick layer on the soil surface. At the sampling dates, the compost layer was removed and only the underlying soil was sampled. Soil pH and total organic carbon content were not affected by the amendments. Soil respiration was significantly higher in compost amended soils compared to the unamended soil except with C4 on day 72. Addition of composts increased plant growth and P uptake was highest on day 72 with C1 and C4. Compost had little effect on the available P concentration, but on day 28, organic P concentrations were lower whereas inorganic P concentrations were higher than in the unamended soil suggesting net mineralisation of organic P. On day 72, the concentrations of the less labile inorganic and organic P forms were higher in the

compost treatments compared to the unamended control suggesting precipitation and fixation as well as synthesis of organic P. This study showed that mulching with composts with high concentrations of available and total P can provide plants with P and also increase soil P concentrations which could reduce the fertilizer requirement for the following crop.

Key words: organic materials; P mineralisation; P pools; plant growth

1. Introduction

Although the total P concentration is high in most soils, P deficiency is the second limiting factor to crop production after N (*Holford*, 1997; *Richardson* et al., 2009). Phosphorus plays a key role in many metabolic pathways and is a structural component of macromolecules such as DNA (*Fuentes* et al., 2008). Soil P can be divided into organic and inorganic forms. The inorganic P in the soil solution, which is readily available to plants, is rapidly converted into less available forms by adsorption and precipitation with Fe and Al ions at low pH, or Ca ions at high pH (*Hinsinger*, 2001; *Khan and Joergensen*, 2009; *Takeda* et al., 2009). To overcome P deficiency, inorganic fertilizers are applied to crops (*Bertrand* et al., 2003; *Evans and Scott*, 2007). However, P fertilizer efficiency in the year of application is only about 20 % (*Bolland and Gilkes*, 1998; *Damodar Reddy* et al., 1999) because fertilizer P is rapidly converted into poorly available forms (*Verma* et al., 2005; *Vu* et al., 2008) which leads to P accumulation in the top soil (*Abad* et al., 2001; *Cornish*, 2009) and may result in particulate P in surface runoff and surface water eutrophication (*Sharpley and Moyer*, 2000; *Song* et al., 2007).

World reserves of rock phosphate which is the basis of inorganic P fertilizers, are limited. At current P fertilizer consumption rates, it is estimated that P reserves with higher P concentration will be depleted in the next 50-100 years (*Isherwood*, 1998; *Smil*, 2000). Upon their depletion, reserves with low P concentration will have to be exploited which increases fertilizer costs. Therefore, alternative sources of P for plants and improved management of native soil P are important for economic and environmental sustainability of agricultural production (*Richardson* et al., 2009).

The effect of organic amendments on P availability depends on the type of organic material, its P concentration, amount added and management practices (*Kwabiah* et al., 2003). Application of organic materials increases P availability and P uptake by plants not only directly by adding P to the soils, but also indirectly through release and production of organic anions and stimulation of microbial activity during decomposition, which can increase P mobilisation and decrease P fixation (*Ayaga* et al., 2006; *Fuentes* et al., 2006; *Khan and Joergensen*, 2009).

Composting is a simple and economic way for organic waste treatment (*Garrido Hoyos* et al., 2002; *Speir* et al., 2004) and application of compost can improve soil physical, chemical and biological properties (*Odlare* et al., 2008; *Uygur and Karabatak*, 2009). The nutrient concentration of the composts depends on the feedstock material that is the organic materials used to produce the compost. Compared to other organic amendments such as plant residues and manure, composts decompose more slowly because as a result of the composting process, they consist of highly decomposed compounds. This may reduce nutrient release but could also increase the length of time during which nutrients are released. The slow and sustained nutrient release may reduce nutrient leaching and supply plants with nutrients throughout growth.

Although there are many studies showing the effect of compost application on soil properties and P availability, little is known about the effect of composts prepared from different feed stocks on P pools and plant P uptake. These composts could differ not only in P concentration but also in decomposability which may affect their impact on soil properties and plant growth.

The aim of this study was to determine the effect of composts derived from different feed stocks on soil P pools and P uptake by wheat, and relate compost properties to the observed effects.

2. Materials and methods

2.1 Experimental design

A loamy sand with low concentration of available P was collected from 0-20 cm depth in a natural bushland in Monarto (latitude $35^{\circ}05^{\circ}$ S, longitude $139^{\circ}06^{\circ}$ E and elevation 166 m), a semiarid region of South Australia (Table 1). The soil was air dried at room temperature and all visible debris was removed manually before sieving to < 5 mm. The soil was filled in 1 kg pots and adjusted to 80 % water holding capacity. Pre-germinated wheat (*Triticum aestivum* cv. Krichauff) seeds were sown before compost application.

Four different composts were used: C1 (from animal manure and straw), C2 (from garden waste), C3 (from wood chips and bark) and C4 (from kitchen waste) (Table 2). The composts were applied as 2.5 cm thick layer on the soil surface corresponding to 135, 150, 130 and 140 g moist compost per pot for C1, C2, C3 and C4, respectively. A further treatment consisted of the unamended soil. Application of compost as mulch is quite common in horticulture and agriculture, particularly in warm

and dry climate because the mulch can reduce water loss from the soil. The pots were kept in a glasshouse under natural light conditions and watered regularly to maintain soil moisture.

Soil and plants were destructively sampled 14, 28 and 72 (grain filling) days after sowing with four replicates per treatment and sampling time. In the compost treatments, the compost layer was removed and only the underlying soil was sampled.

2.2 Analysis of soil and composts

Soil pH and EC were measured in a 1:5 soil to water suspension (w/w) after 1 hour shaking at 25°C. Water holding capacity of the soil was measured using a sintered glass funnel connected with a 100 cm water column (ψ m = -10 kPa). Total organic carbon (TOC) was measured by the *Walkley* and *Black* method (1934).

For determination of soil respiration, 30 g soil was filled in cores of 3.7 cm diameter and 5 cm height with a nylon mesh base and adjusted to a bulk density of 1.5 g cm⁻³. The cores were placed in 1 L glass jars with air-tight lids which were incubated in the dark at 22-25°C for 21 days. Headspace gas was withdrawn daily and the CO₂ concentration measured (t1) with an IR CO₂ gas analyser (Servomex 1450 Food Package Analyser, Crowborough, UK). After measurement, the jars were opened and flushed with ambient air. After closing, the CO₂ concentration was determined (t0). The relationship between gas analyser reading and CO₂ concentration was determined by injecting known amounts of CO₂ into empty glass jars of similar size. The CO₂ evolved in the measurement interval was calculated as the difference between t1 and t0

multiplied by the volume of the jars. During incubation, soil moisture was maintained by weight with autoclaved reverse osmosis water.

Available P in the soil and composts was determined using anion exchange resin membranes as described in Kouno et al. (1995). In each replicate, soil P pools were assessed in three separate subsamples. The first subsample was used to determine water-soluble P where 2 g soil was shaken for 16 h in 30 ml reverse osmosis (RO) water after which the P concentration in the filtrate was measured as described below. Microbial P was measured in the second subsample as described by Kouno et al. (1995). The third subsample was used for sequential P fractionation *(Hedley et al., 1982;*) *Tiessen and Moir*, 1993) where first labile and then less labile P forms are extracted by shaking the soil sequentially with different extractants. One g soil was sequentially extracted (at a 1:30 soil: solution ratio) with 0.5 M NaHCO₃, 0.1 M NaOH, 1 M HCl and again 0.1 M NaOH. The second extraction with NaOH was carried out because substantial amounts of NaOH-soluble P could be released after removal of HCl-soluble P from this soil. Extraction with concentrated HCl was not included because preliminary tests had shown that in this soil no P was detected in this fraction. Five mL of the extract from the NaHCO₃ and NaOH fractions was acidified to pH 1.5 to precipitate organic matter, centrifuged and the supernatant was analysed for inorganic P (Pi). Total P in NaHCO₃ and NaOH fractions was analysed as described by Huang and Zhang (2004). Briefly, 5 mL extract was digested with 1 mL 5% potassium persulphate at 90°C for 16 h and was analysed for total P. Organic P (Po) was calculated as the difference between total P and inorganic P. Residual P was determined by digesting the residual soil in HNO₃-HClO₄ (6:1). The total P concentration in composts and soil was also determined by wet digestion with a mixture of nitric acid and perchloric acid (6:1). The P concentration in all fractions was measured colorimetrically according to *Murphy* *and Riley*, (1962) at a wavelength of 712 nm after neutralization when necessary. Water-soluble and bicarbonate-extractable (NaHCO₃) Pi and Po are considered labile pools whereas NaOH-extractable Pi and Po, acid-extractable (HCl) Pi and residual P are less labile pools (*Hedley* et al., 1982; *Tiessen and Moir*, 1993).

2.3 Plant analysis

Shoots were cut at the soil surface and rinsed with water to remove dust. The roots were collected from the soil and washed with reverse osmosis water. Shoots and roots were oven dried at 65° C for three days. Oven-dried shoots were ground and digested with a mixture of nitric acid and perchloric acid (6:1). The P concentration was measured with molybdate-vanado-phosphate method at 390 nm (*Hanson*, 1950). Shoot P uptake was calculated by multiplying P concentration with shoot dry weight.

2.4 Statistical analysis

The experiment was completely randomized block design with two factors (five treatments and three sampling dates) and four replicates. The data was subjected to two-way analysis of variance (GenStat for Windows 11.0). Tukey test was used to determine significant differences among treatments. The least significant difference (LSD) was calculated at 5 % of probability. Only significant differences are mentioned in the text ($P \le 0.05$).

3. Results

3.1 Compost characterization

The four composts differed in a number of properties (Table 2). Compost 1 was medium-textured and had the highest pH, available P concentration and a low total C

concentration and C/P ratio. Compost 2 was fine-textured and had a low available P concentration and the lowest C/N ratio, but a high total P concentration. Compost 3 was coarse textured, had the lowest pH and available P and total P concentrations, but the highest total C concentration, C/P and C/N ratios. Compost 4 was medium-textured and characterised by high EC and high P availability; it had the highest total P concentration and the lowest C/P and C/N ratios.

3.2 Soil properties

The compost mulch had no effect on the pH of the underlying soil (data not shown). Mulching with C1, C2 and C4 increased the electrical conductivity (EC) of the underlying soil compared to the unamended soil, but the $EC_{1:5}$ remained below 0.7 dS m⁻¹ which is considered as threshold level above which growth of many crop plants can be affected (data not shown) (*Rengasamy* et al., 1984; *Tillman and Surapaneni*, 2002). The soil organic carbon concentration did not differ significantly among treatments and did not change over time.

In all treatments, cumulative respiration was highest in the samples taken on day 14 which can be explained by high temperature during the incubation period for this sampling date. To allow comparison between treatments across all sampling dates, cumulative respiration is presented in percentage of the unamended soil for each sampling time (Fig. 1). At all three sampling times cumulative respiration was higher in soils mulched with C1, C2 and C3 than in the unamended soil whereas C4 increased cumulative respiration only on days 14 and 28.

3.3 Soil P pools

3.3.1 Changes in P pools in the unamended soil over time

There were no significant changes in the concentrations of microbial P and NaHCO₃-Po in the unamended soil over time (Table 3). From day 0 to day 14, the water-soluble P concentration significantly increased and the NaOH-Pi and Po concentrations decreased. Compared to day 0, the concentrations of NaHCO₃-Pi and HCl-P were significantly lower on day 72 whereas the NaOH-Po and residual P concentrations were higher. The NaOH-Pi concentration decreased significantly from day 0 to day 28 and then remained stable.

To analyse the effect of compost addition on soil P pools, the soil P pool concentrations in the amended soils are presented after subtracting the concentrations in unamended soils. Compared to the unamended soil, the compost mulch increased soil P pool concentration with greater increases in the labile P pools than the less labile P pools.

3.3.2 Changes in P pools in amended soils

On day 14, mulching with C1 increased the concentrations of all P pools compared to the unamended soil except microbial P, but particularly the concentrations of NaHCO₃-Po, NaOH-Pi and HCl-P whereas the increases in water-soluble P, NaHCO₃-Pi and residual P were small (Fig. 2). On day 28, there was a strong increase in the concentrations of water-soluble P, NaHCO₃-Pi, NaOH-Pi and HCl-Pi compared to the unamended soil, whereas the concentrations of the organic P pools (NaHCO₃-Po and NaOH-Po) and HCl-P decreased. The sum of increases was greater than that of decreases indicating P release from the compost. On day 72, the concentration of NaOH-Po strongly increased compared to the unamended soil whereas the concentrations of the other pools were little affected except for a decrease in the concentration of NaHCO₃-Pi.
The other composts induced smaller changes in P pool concentrations compared to the unamended soil than C1 (Fig. 2 B, C and D). On day 14, mulching with C2 resulted in higher concentrations of NaOH-Po and residual P compared to the unamended soil whereas the concentration of the other pools was little affected (Fig. 2 B). On day 28, the concentration of NaOH-Po with C2 mulch was lower than in the unamended soil, but the concentration of the other pools was not affected. On day 72 the concentration of NaOH-Pi was increased by mulching with C2 whereas that of residual P was decreased. Mulching with C3 had no effect on P pool concentrations in the underlying soil on day 14 (Fig, 2 C). But C3 resulted in a decrease in the concentration of NaOH-Po on day 28 and an increase in NaOH-Pi on day 72 compared to the unamended soil. On day 14, mulching with C4 increased the concentration of NaOH-Po, HC1-Pi and residual P were higher in the soil mulched with C4 than in the unamended soil.

In general, the composts had little effect on the P pool concentrations on day 14 except for an increase in the concentration of NaOH-Po by C1, C2 and C4. On day 28, all composts induced a decrease in NaOH-Po concentration and on day 72, the concentrations of the less labile P pools (NaOH-Pi and NaOH-Po) were higher than in the unamended soil.

3.4 Plant dry matter and shoot P uptake

In all treatments, shoot dry matter increased 8-25 fold from day 28 to day 72, and root dry matter increased 2-5 fold with a greater increase in the compost amended soil than in the unamended soil (Table 4). Shoot and root dry matter did not differ significantly

between treatments on days 14 and 28. Compared to the unamended soil, shoot dry matter on day 72 was highest in C4 (4 fold) and C1 (2 fold) amended plants, whereas root dry matter was highest with C4.

On day 14, compost addition increased shoot P concentration compared to the unamended plants. Shoot P concentration decreased in all treatments over time. On day 72, shoot P concentration was lowest with C3 and highest with C1; compared to the unamended soil, only C1 significantly increased shoot P concentration (Table 4). Shoot P uptake increased over time and was greatest on day 72 (Fig. 3). On days 14 and 28, P uptake was not affected by compost addition but on day 72, P uptake was increased by all composts with the greatest increase with C1 and C4.

4. Discussion

This study showed that the effect of composts on plant growth, P uptake and soil P pools varies with compost type, but there were some general trends in the soil P pools that occurred with all composts.

Plant growth was slow, therefore the changes in the soil P pools between day 0 and day 28 were mainly driven by P input from the compost mulch, whereas in the period from day 28 to day 72, plant P uptake could also have played a role.

4.1 General effects of composts

Although the compost mulches did not increase total organic carbon content of the underlying soil, they increased cumulative respiration significantly except for C4 on day 72. This increase can be explained by leaching of soluble carbon from the compost mulch into the underlying soil (as a result of the watering of the pots). The strong decrease in cumulative respiration over time with C4 may be due to the easily decomposable nature of the feedstock (kitchen waste) and its fine texture. Although the composts were placed on the soil surface, some decomposition will have occurred over the 72 days by compost microbes and soil microbes colonising the compost mulch. The C4 compost would be decomposed more rapidly than the other composts which would reduce leaching into the soil over time.

All composts increased plant growth compared to the unamended soil which indicates that nutrients released by the composts were taken up by the plants and enhanced their growth (*Butler and Muir*, 2006; *Dou* et al., 2009). On day 72, root and shoot growth were greatest with C4 which may be explained by the nutrient-rich feedstock (kitchen waste) as evident in the low C/P and C/N ratios.

On day 72, all composts increased plant P uptake with the greatest increase by mulching with C1 and C4. This could be due to the higher amount of available P added as well as the greater root biomass with composts which would increase the soil volume exploited (*Sinaj* et al., 2002). The concentration of the labile P pools was not affected by compost application compared to the unamended soil or even decreased in the case of C1. However, it is likely that some P was released from the compost mulch, entered the labile P pools and was taken up by the plants.

4.2 Effect of composts on P pools

Through leaching resulting from the watering of the pots, the compost mulch served as a continuous P source throughout the experiment. This explains why even on days 28 and 72, there was strong net increase in P pools in the compost treatments although the plants took up P in this period.

On day 28, there was a conversion of organic P into inorganic P in the compost treatments suggesting net mineralisation of organic P which was probably enhanced by the stimulation of microbial activity by the composts. But on day 72, the concentration of the less labile inorganic and organic P forms was higher in the compost treatments compared to the unamended control suggesting precipitation and fixation as well as synthesis of stable organic P (*Garcia-Gil* et al., 2000; *Park* et al., 2004; *Shen* et al., 2011). This net increase in the concentration of the less labile P pools indicates that more P was released from the composts than was taken up by the plants. Although considered to be less labile, NaOH-P and HCl-P can be depleted in the rhizosphere (*Rose* et al., 2010; *Mat Hassan* et al., 2012) indicating that they are plant available. This suggests that composts can be a long-term and sustainable P source for plants.

Compared to the unamended soil, mulching with C1 resulted in the strongest increases in soil P pool concentrations on days 14 and 28 which can be explained by the high amount of available P added in this compost and its low C/P ratio. This is in agreement with other studies which found an immediate increase in available P with application of organic materials with high P concentrations (*Iyamuremye* et al., 1996; *Nziguheba* et al., 1998; *Odlare* et al., 2008). Mulching with C3 had no effect on soil P pools on day 14 which can be explained by its low concentrations of available P and total P. This compost is made from wood chips and has a very high C/P ratio which could result in net immobilisation within the compost (*Singh* and *Amberger*, 1998;

Uygur and *Karabatak*, 2009). However later, some of the compost P was leached into the soil as indicated by the net increase in the size of the P pools and by plant P uptake.

It should be noted that the increase of the less labile P forms may also directly derived from the composts through leaching of poorly labile P. The increase in the concentration of the less labile P pools in the soil mulched with C2 on days 28 and 72 was accompanied by a decrease in residual P which suggests conversion of residual P into other less labile P pools (*Aulakh* and *Pasricha*, 1991; *Wang* et al., 2007).

Among the composts, mulching with C1 and C4 increased plant growth and net soil P pool concentrations most whereas the effect of C3 was small. Thus composts with high concentrations of available and total P can provide plants with P and also increase soil P concentrations which could reduce the fertiliser requirement for the following crop.

5. Conclusion

This study showed that mulched composts can act as slow release fertilisers supplying the plants with P and other nutrients particularly in the later stages of growth. Among the composts, mulching with C1 and C4 increased plant growth and net soil P pool concentrations most whereas the effect of C3 was small. Thus composts with high concentrations of available and total P can provide plants with P and also increase soil P concentrations which could reduce the fertiliser requirement for the following crop.

Acknowledgements

This study was funded by the Australian Research Council.

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Table 1 Soil properties

Properties	Unit	Soil
Sand	%	82.5
Silt	%	10
Clay	%	7.5
pН		8
Electrical conductivity	dS m ⁻¹	0.1
Total organic carbon	g kg ⁻¹	9
Available P	mg kg ⁻¹	5.3
Total P	mg kg ⁻¹	170
Total N	g kg ⁻¹	0.9
Bulk density	g cm ⁻³	1.5

Properties	Units	C1	C2	С3	C4
Main feed stock		Animal manure	Garden	Wood	Kitchen
		and straw	waste	chips and	waste
				bark	
pH		8.6	8.1	6.4	7.1
Electrical conductivity	dS m ⁻¹	0.8	4.2	1.0	7.5
Total organic carbon	g kg ⁻¹	119	178	289	118
Available P	mg kg ⁻¹	27.9	3.8	1.6	11.7
Available P added	mg pot ⁻¹	3.9	0.5	0.2	1.6
Total P	mg g ⁻¹	1.7	2.4	0.2	5.7
Total P added	mg pot ⁻¹	240	338	28	803
C/P ratio		81	104	1087	17
C/N ratio		11	9	35	12
Particle size					
4.75-3.25 mm	%	37	26	35	34
2-1 mm	%	46	42	56	53
0.5-0.25 mm	%	17	32	9	13

Table 2 Properties of composts (C1, C2, C3 and C4)

Table 3 Concentration of P pools in the unamended soil on days 0, 14, 28 and 72 (n=4). Values in the same row followed by different letters are significantly different ($P \le 0.05$).

Day	0 14		28	72		
P pools	(mg kg ⁻¹)					
Water-soluble P	6.1 a	13.4 b	10.3 b	5.5 a		
Microbial P	3.9 a	5.2 ab	4.2 a	3.7 a		
NaHCO ₃ -Pi	10.6 b	9.9 b	9.22ab	6.6 a		
NaHCO ₃ -Po	2.9 a	5.8 a	6.6 a	2.1 a		
NaOH-Pi	31.6 c	24.8 b	16.7 a	19.6 ab		
NaOH-Po	38.1 b	25.7 a	33.7 b	71.0 c		
HC1-P	20.5 b	21.8 b	19.5 ab	17.5 a		
Residual P	26.5 a	28.0 a	34.2 b	52.4 c		

Table 4 Wheat shoot and root dry matter and shoot P concentration on days 14, 28 and
72 in unamended soils (C) and soils amended with composts (C1-C4) (n=4). The LSD
refers to $P \le 0.05$.

	Shoot dry weight		Root dry weight		P concentration				
	(g pot ⁻¹)			(g pot ⁻¹)			(g kg ⁻¹)		
Day	14	28	72	14	28	72	14	28	72
Treatments									
С	0.10	0.21	1.64	0.16	0.22	0.42	3.94	2.68	1.78
C1	0.10	0.25	4.57	0.14	0.12	0.61	4.37	3.23	2.12
C2	0.08	0.25	3.40	0.09	0.15	0.51	5.07	2.20	1.97
C3	0.09	0.25	2.92	0.08	0.19	0.63	4.47	1.42	1.45
C4	0.08	0.25	6.15	0.08	0.25	0.96	4.66	2.49	1.66
LSD (5 %)		0.27			0.14			0.41	



Fig. 1 Cumulative respiration after 21 days in percentage of the unamended soils in the soils amended with composts (C1-C4) sampled on days 14, 28 and 72 (n=4). Vertical lines indicate standard deviation. The horizontal line represents the unamended soil (100 %).



Fig. 2 Concentration of P pools on days 14, 28 and 72 in soils amended with composts (C1-C4) after subtracting the concentrations of the unamended soil. Negative values indicate lower concentrations than the unamended soil, positive values higher concentrations.



Fig. 3 Shoot P uptake of wheat grown in unamended soils (C) and soils amended with composts (C1-C4) on days 14, 28 and 72 (n=4). Vertical lines on the bars indicate standard deviation. The single thick line shows the least significant difference.

Chapter 3

Mobilisation of rock phosphate by surface application of compost

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The work contained in this chapter is published in Biology and Fertility of Soils

Verma SL, Penfold C, Marschner P (2013) Mobilisation of rock phosphate by surface application of compost. Biol Fertil Soils 49: 287-294

STATEMENT OF AUTHORSHIP

Mobilisation of rock phosphate by surface application of compost

Biology and Fertility of Soils (2013): DOI 10.1007/s00374-012-0717-y

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NOTE:

This publication is included on pages 50-58 in the print copy of the thesis held in the University of Adelaide Library.

It is also available online to authorised users at:

http://dx.doi.org/10.1007/s00374-012-0717-y

Chapter 4

Compost incorporation increases phosphorus release from rock phosphate

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Compost incorporation increases phosphorus release from rock phosphate

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Abstract

It has been suggested that the combined application of compost with rock phosphate (rock P) may improve solubility of rock P, but it is not clear if this is also the case in neutral and near neutral soils. A 75-day glasshouse study was conducted in a loamy sand soil (pH_{1:5} 7.7) with low P availability. Two different composts (C1 from garden waste, fine textured and C2 from wood chips, coarse textured) were applied as mulch or incorporated into the soil into which rock phosphate was mixed at 35 mg total P kg⁻¹. Other treatments included composts alone or rock P alone as well as an unamended soil. Wheat was sown and harvested at maturity (day 75). The concentration of different P pools was analysed on days 0 and 75. Plant dry matter and P uptake were highest with incorporation of C1 with or without rock P, whereas P concentration was highest with incorporated C2. Rock P alone did not affect the soil properties but incorporation of composts with or without rock P increased pH, electrical conductivity and total organic C. Incorporation of C1 increased the concentrations of NaHCO₃-Pi, NaOH-Pi and HCl-P from day 0 to day 75 whereas the concentrations of labile P pools (resin P and microbial P) and organic P fractions of the NaHCO₃ and NaOH pools decreased which suggests that compost addition induced P flux between soil P pools. The incorporated composts mobilised rock P because the measured increases in P pool concentrations between incorporated compost alone and compost with rock P were greater than the sum of rock P alone and compost alone. On the other hand, compost mulches had a smaller effect on the measured parameter and did not mobilise rock P. In conclusion, incorporation of fine-textured compost increases P availability by three processes: (i) addition of P with compost, (ii) inducing fluxes between soil P pools and, (iii) when rock P is added to the soil, mobilising P from rock P.

Key words: inorganic fertilizer, organic materials, P solubility, sequential P fractionation.

Introduction

Soil phosphorus (P) management is a challenge due to the low concentration of available P in most soils and the low use efficiency and high cost of inorganic fertilisers. Low soil P availability is the result of conversion of soluble P into insoluble Al and Fe phosphates in acidic soils and into Ca phosphate in neutral to alkaline soils (Hinsinger 2001; Bertrand et al. 2003). Only 10-20 % of applied P is taken up by the plants in the year of application because the applied soluble P is rapidly converted into poorly available forms which accumulate in soils (Bolland and Gilkes 1998; Guo et al. 2000; Vu et al. 2008). Rock phosphate is main source for manufacturing of P fertilisers and high grade rock phosphate reserves may be exhausted in 50 to 100 years (Steen 1998). Manufacturing of P fertilisers also requires sulphuric or phosphoric acids which are very expensive, these costs in addition to high energy prices are responsible for the increasing fertiliser costs (Easterwood et al. 1986). Because of the low P fertiliser efficiency, P application rates often exceed plant demand which may result in the surface water eutrophication (Maftoun and Moshiri 2008). The diminishing P reserves and high manufacturing cost of chemical fertiliser have led to increasing interest in the use of alternative P sources like compost, manure and low grade rock phosphate. Rock phosphate solubility increases with decreasing pH, therefore it is soluble in acidic soils (Hongqing et al. 2001) but not in neutral and alkaline soils (Bangar et al. 1989).

The effectiveness of rock P can be increased by combining rock P with organic amendments or co-composting of rock P with organic wastes (Reddy 2007; Pramanik et al. 2009), partial acidulation of rock P (Hammond et al. 1980) and use of P-solubilising microbes (Azcon et al. 1976; Goenadi and Sugiarto 2000). Depending on the decomposability of organic wastes, 25-60% of rock P can be converted into soluble P forms (Singh and Amberger 1990) through organic acids and protons produced during the decomposition of organic material (either in the soil or during composting). While this has been shown when rock P was added with plant residues and green manure in the soil or during composting (Mishra and Banger 1984; Zaharah and Bah 1997), it is not clear if compost influences rock P availability when both are added to soil. Compared to the residues and green manure, compost decomposability is lower. Thus, they may not generate enough organic acids and protons to increase P release from rock P, particularly if they are applied as mulch instead of being incorporated. Compared to compost mulching, incorporation of compost has a greater effect on nutrient availability and soil physical properties (Cogger et al. 2008). The compost effect may also be modulated by the particle size of the compost because it affects decomposition rate; coarse textured composts have lower decomposition rates than fine textured composts (Duong et al. 2011). Further, it is not clear if compost addition to rock P can increases P release when applied to a soil with a neutral to slightly alkaline pH where the solubility of rock P alone is very low (Banger et al. 1989). The main objectives of this study were to determine the effect of (i) compost type (fine or coarse texture) and (ii) application method (mulched or incorporated) on the solubility of incorporated rock P and P uptake by wheat.

Materials and Methods

Experimental setup

A loamy sand soil with low available P concentration was collected from 0-20 cm depth in a natural bushland in Monarto ($35^{\circ}05$ 'S and $139^{\circ}06$ 'E, elevation 212 m). The soil was air-dried and passed through a 5 mm sieve. The properties of the soil are as follows: pH_{1:5} 7.7; EC_{1:5} 0.05 dS m⁻¹; sand 82.5%; clay 7.5%; silt 10%; total organic C 8 g kg⁻¹; resin P 5.1 mg kg⁻¹; total P 179 mg kg⁻¹, total N 0.9 g kg⁻¹ and bulk density 1.6 g cm⁻³. The soil was filled in 1 kg pots and adjusted to 75% water holding capacity. Previous experiments had shown that this water content was optimal for plant growth and microbial activity in this soil (Setia et al. 2011).

Two different composts were used in this experiment: C1 (from garden waste, fine textured) and C2 (from wood chips and barks, coarse textured) (Table 1). The rock P Nutriphos Guano Powder, fossilised seabird dropping with a combination of slow-release and soluble P (RP) containing total P 139 g kg⁻¹, water-soluble P 13.9 g kg⁻¹ and citrate-soluble P 14.4 g kg⁻¹ was incorporated into the soil at the rate of 0.25 g kg⁻¹ soil (equivalent to 35 mg total P kg⁻¹ soil). This rate was based on a preliminary experiment where it resulted in a small increase in the concentration of available P. The composts were applied as mulch (2.5 cm thick layer, corresponding to 156 g pot⁻¹ for C1 and 130 g pot⁻¹ for C2) on the soil surface or the same weight of the compost was incorporated in the soil with or without rock P. Further treatments included soil with rock P alone and the unamended soil. Wheat (*Triticum aestivum* cv. Krichauff was sown and harvested at maturity (day 75).

Soil and Compost Analyses

Soil particle size was analysed by the hydrometer method (Ashworth et al. 2001). Soil pH and EC were measured in a 1:5 soil to water suspension (w/w) after shaking 1 hour at 25°C. The soil water holding capacity was measured using a sintered glass funnel connected with a 100 cm water column (ψ m = -10 kPa). Total organic carbon (TOC) was measured by the Walkley and Black method (Walkley and Black 1934).

At sampling on days 0 and 75, the compost mulch was removed before the underlying soil was sampled for analysis. The compost mulch was analysed for resin, microbial and total P.

For soil respiration determination, 20 g soil sampled on days 0 and 75 was added to cores of 3.7 cm diameter and 5 cm height which were then placed in 1 L glass jars with air tight lids and equipped with a septum. The cores were incubated in the dark at 22-25 °C for 21 days. Daily, headspace gas was withdrawn and the CO₂ concentration measured with an infrared CO₂ gas analyser (t1) (Servomex 1450 Food Package Analyser, Crowborough, UK). After measurement, the jars were opened and flushed with ambient air. After closing, the CO₂ concentration was determined (t0). The CO₂ evolved in the sampling interval is the difference between CO₂ concentration at t1 and t0 multiplied by the gas volume of the jars. The relationship between detector reading and CO₂ concentration was determined by injecting known amounts of CO₂ injected into empty glass jars of similar size. During incubation, soil moisture was maintained by weight with autoclaved reverse osmosis water.

Available P and microbial P in the soil and composts were determined using anion exchange resin membranes as described in Kouno et al. (1995). Microbial P was calculated from the difference between samples with or without hexanol (Kouno et al. 1995). Total P in the soil and composts was determined by wet digestion with a mixture of nitric acid and perchloric acid (6:1). Soil samples from days 0 and 75 were analysed by sequential P fractionation based on Hedley et al. (1982) and Tiessen and Moir (1993) with some modifications. One g of soil was placed in 50-ml centrifuge tube and was sequentially extracted (at a 1:30 soil: solution ratio) with water, 0.5 M NaHCO₃, 0.1 M NaOH, 1 M HCl and again 0.1 M NaOH. Five mL of the extract from the NaHCO₃ and NaOH fractions was acidified to pH 1.5 to precipitate organic matter, centrifuged (1610 \times g) and the supernatant was analysed for inorganic P. Total P in NaHCO₃ and NaOH fractions was analysed by the method described by Huang and Zhang (2009). Briefly, 5 mL extract was digested with 1 mL 5% potassium persulphate at 90°C for 16 h and was analysed for total P. Organic P was calculated as the difference between total P and inorganic P. Residual P was determined by digesting the residual soil in nitric acid and perchloric acid (6:1). The P concentration in all fractions was measured at a wavelength of 712 nm after neutralization if necessary (Murphy and Riley 1962). Water-soluble and bicarbonate-extractable (NaHCO₃) Pi and Po are considered as labile pools whereas NaOH-extractable Pi and Po, acid-extractable (HCl) Pi and residual P are less labile pools.

Plant analysis

Shoots were cut at the soil surface and rinsed with reverse osmosis water to remove soil. In mulched compost treatments, shoots were cut after removing compost layer. The roots were collected from the soil and washed with water. Shoots were oven dried at 60°C for three days, finely ground and then 250 mg shoot material was digested with a mixture of nitric acid and perchloric acid (6:1). The P concentration in the digest was measured with molybdate-vanado-phosphate method at 390 nm (Hanson 1950). Shoot P uptake was calculated by multiplying P concentration with shoot dry weight.

Statistical analysis

The experiment was completely randomized block design with two factors (six treatments and two sampling dates) and four replicates. The data was subjected to two-way analysis of variance (Genstat for Windows 11.0). Linear regression and Tukey test was used to determine correlation and significant differences of treatments. The least significant difference (LSD) was calculated at 5 % of probability. Only significant differences are mentioned in the text ($P \le 0.05$).

Results

Compost properties

Compared to C2, C1 had a higher pH, EC, total P and a finer texture, but lower concentrations of total organic C and available P and lower C/P and C/N ratios (Table 1). The amount of available P added to the soil was similar for both composts, but more total P was added with C1 compared to C2. From day 0 to day 75, total P and microbial P concentration of both compost mulches decreased, whereas the resin P concentration increased with a greater increase in C2 than C1 (Table 2).

Soil properties

Irrespective of rock phosphate addition, the incorporation of C1 increased the soil pH from 7.7 to 8.4 whereas soil pH decreased from 7.7 to 7.1 with incorporation of C2 (data not shown). The soil electrical conductivity significantly increased with incorporation of composts with and without rock phosphate with greatest increase in C1 where the EC ranged from 0.13 to 0.30 dS m⁻¹ compared to 0.05 dS m⁻¹ in the unamended soil. Compost incorporation with or without rock phosphate increased the total organic C concentration at days 0 and 75 with the greatest increase with C1. Cumulative respiration was higher for soils sampled on day 0 than for soils sampled on day 75 (Fig. 1). Compost incorporation increased cumulative respiration compared to the unamended soil at both sampling dates with greater increases by C1 than C2. Compared to the unamended soil, mulching of composts had no effect on pH, EC or total organic C of the soil. Cumulative respiration for samples taken on day 0 was not affected by mulched composts, but cumulative respiration of the samples taken on day 75 was increased by compost mulches.

Changes in soil P pools over time

In the unamended soil, the concentrations of water-soluble P, microbial P, NaOH-Pi and residual P increased from day 0 to day 75, whereas the concentrations of NaHCO₃-Pi and NaOH-Po decreased (Table 3).

Compared to the unamended soil, incorporation of composts with or without rock phosphate increased the concentrations of most measured P pools whereas mulching

had little effect. With incorporated compost the effect was greater with C1 than C2 (Fig. 3). Only the residual P concentration on day 75 was lower in the amended soils compared to the unamended control. Rock phosphate alone had no effect on the concentration of the labile P pools, but increased the concentration of NaOH-P and HCl-P compared to the unamended soil.

Among labile P pools, the concentration of water-soluble P, microbial P and NaHCO₃-Po significantly decreased from day 0 to day 75 with incorporation of C1 with or without rock P whereas the NaHCO₃-Pi concentration significantly increased only in C1 with rock P. With C2 the concentration of labile P pools remained unchanged over time except for an increase in microbial P concentration when rock P was added. For the less labile P pools, the concentrations of NaOH-Pi and HCl-P significantly increased from day 0 to day 75 with mulched C1 with rock P and incorporation of C1 with or without rock P whereas the concentration of NaOH-Po and residual P decreased with incorporation of C1 with or without rock P. The concentration of NaOH-Pi significantly increased from day 0 to day 75 with incorporated C1 with rock P but decreased with incorporation of C2 and reverse was true for NaOH-Po. The HCl-P concentration significantly increased over time with mulched C2 with rock P (Fig. 3).

To assess if the combination of compost with rock P resulted in release of P from rock P, the concentration of a given pool with compost alone and with rock P alone were added to give an expected value. If the concentration measured in the treatment of compost with rock P is greater than the expected value, rock P was mobilised. With incorporated composts, the increase in the concentrations of water-soluble P, NaHCO₃-Po, NaOH-Pi and Po and HCl P on day 0 between the composts alone and composts with rock P was greater than expected (Fig. 3). On day 75, the measured concentration was greater than the expected concentrations of NaHCO₃-Pi

and Po and NaOH-Pi but measured and expected values did not differ for water-soluble P, microbial P, NaOH-Po and HCl-P concentrations. Measured and expected values did not differ with mulched composts.

Plant growth and P uptake

After 75 days, wheat shoot dry weight was significantly higher in all treatments with C1 than the unamended soil, whereas C2 had no effect on shoot dry weight or decreased it (Fig. 2). Root dry weight was increased only with incorporated C1 with or without rock P. Compared to the unamended soil, the shoot P concentration was increased only with incorporation of C2 with or without rock P whereas shoot P uptake was increased with all C1 treatments. The higher P concentration with incorporated C2 can be explained by lower plant dry weight with this treatment. Shoot P uptake was positively correlated with total P added with amendments ($r^2 = 0.80$, P ≤ 0.01). Rock P alone did not affect plant growth and P uptake.

Discussion

This study showed that compost incorporation resulted in a greater increase in plant growth and P uptake as well as soil P pools than mulched compost. Incorporated compost also appeared to have increased the solubility of rock P.

Effect of composts on soil properties and plant growth

The changes in soil pH and EC induced by the incorporation of the composts can be explained by the pH and EC of the composts (Cox et al. 2001; Larney et al. 2008): compared to C2, C1 had a higher pH and EC and consequently resulted in higher soil pH and EC (Table 2). But the EC remained below 0.7 dS m⁻¹ which is threshold level above which growth of most crop plants can be affected (Rengasamy et al. 1984; Tillman and Surapaneni 2002). The increase in total organic C following compost incorporation can be explained by the addition of organic matter with composts (Ros et al. 2003; Ros et al. 2006). Mulched compost on the other hand had little effect on soil organic C which suggests there was little transfer of carbon from the compost into the underlying soil. This is also supported by the small increase in soil respiration induced by the mulched composts compared to the incorporated composts (Fig. 3).

For soil sampled on day 0, soil respiration was higher with incorporation of C1 with or without rock P than with C2, although the total organic C added with C1 was lower than C2, however C1 was fine textured and had lower C/N and C/P ratios and was therefore more easily degradable. The smaller compost particles result in a larger surface area to volume ratio which increases the accessibility of microbes and can also increase release of nutrients (Bending and Turner 2009).

The differences in particle size and C/N, C/P ratios can also explain the greater effect on plant growth and P uptake by C1 compared to C2. Compared to the unamended soil, C1 increased shoot growth more when incorporated than when mulched whereas the reverse was true for C2. This suggests that for C1, some nutrients were leached from the mulch into the underlying soil over the course of the experiment but the amount was smaller than those available to the plants when the compost was incorporated. The decrease in plant growth with incorporated compared to mulched C2 suggests that the compost contains phytotoxic compounds which are released during decomposition in the soil (Keeling et al. 1994). Further, the high C/N and C/P ratio could also result in N and P immobilisation although this was not evident in the microbial P concentration (Fig. 2) which was lower for C2 than C1.

Soil P pools

Although similar amounts of available P were added with the two composts, incorporation of C1 resulted in greater increases in the concentration of the labile P pools than C2 even on day 0. This was also the case on day 75 except for water-soluble P. The stronger increase on day 0 by C1 suggests that upon incorporation, some previously unavailable P was released, possibly by breaking up of particles or clumps. Since C1 had a higher total P concentration and was finer textured than C2, this release of P would have been greater with C1. The greater increase in labile and non-labile P concentrations on day 75 with C1 can be explained by the mineralisation and solubilisation of compost P over time. Since plant P uptake was also higher with C1, it can be assumed that more P was released than the increase in soil P pools suggests.

Incorporation of C1 also increased the concentration of the non-labile P pools more than C2 on both sampling dates. The short time between compost application and sampling on day 0 (a few hours) suggests that any changes in soil P pools induced by the composts are due to P pools already present in the compost. Thus, the increase on day 0 suggests that the concentrations of NaOH-P and HCl-P were higher in C1 compared to C2 (Table 2). The increase in the concentrations of NaHCO₃-Pi, NaOH-Pi and HCl-P from day 0 to day 75 with C1 can be explained by two processes (i) release of P from the compost (Iyamuremye et al. 1996; Nziguheba et al. 1998; Odlare et al. 2008), and (ii) flux of P among soil P pools (Sanchez and Beck 1994). Although the decreases in the labile and possibly also of the less labile P pools could be explained by plant P uptake, some of the water-soluble P and microbial P appears to have been converted into less labile P pools. Similarly, the decrease in the concentration of NaHCO₃-Po and NaOH-Po suggests mineralisation of organic P. Butler and Muir (2006) and Dou et al. (2009) also reported the release of organically bound P and N after application of compost. Further, compared to the unamended soil, there was a decrease in residual P over time. Thus, although this pool is considered to be poorly available (Schoenau et al. 1989), P was released from this pool and converted into other soil P pools and may also be taken up by the plants directly or after conversion into labile P pools.

The incorporated composts also appear to have mobilised rock phosphate P because the measured increases in the concentrations of labile and non-labile P pools induced by incorporated compost with rock P was greater than the sum of rock P alone and compost alone. This mobilisation could be due to increased microbial solubilisation or solubilisation of rock P by organic acids released during compost decomposition (Zaharah and Bah 1997; Singh and Amberger 1998). An increase in solubility of rock P was also found by Akande et al. (2005) and Kolawole and Tian (2010) with combination of poultry manure and residues with rock P. Further, mobilisation of rock P by the plants when combined with compost can also not be ruled out because the plants grew better than with rock P alone.

Conclusion

This study showed that incorporation of fine-textured compost can not only improve plant growth and increase the concentration of labile and non-labile soil P pools, but it
can also result in mobilisation of rock P. The increased concentrations of non-labile P pools compared to the unamended soil even 75 days after compost incorporation suggest that compost may have longer lasting effect on P availability. The lower concentration of residual P on day 75 in all amended treatments compared to the unamended soil suggests that this P pool is not as stable as generally assumed. Compared to compost incorporation, mulching had little effect on soil P pools and plant growth and did not mobilise rock P.

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Properties	Unit	C1	C2
Main feed stock		Garden waste	Wood chips and bark
рН		8.2	6
Electrical conductivity	dS m ⁻¹	4.3	1
Total organic carbon	g kg ⁻¹	121	168
Available P	mg kg ⁻¹	66	94
Available P added	mg pot ⁻¹	10	12
Total P	g kg ⁻¹	2.2	1
Total P added	mg pot ⁻¹	343	130
C/P ratio		77	118
C/N ratio		14	28
NaHCO ₃ -Pi	mg kg ⁻¹	315	269
NaHCO ₃ -Po	mg kg ⁻¹	43	23
NaOH-Pi	mg kg ⁻¹	78	37
NaOH-Po	mg kg ⁻¹	44	12
HC1-P	mg kg ⁻¹	1199	365
Residual P	mg kg ⁻¹	396	125
Particle size			
4.75-3.25 mm	%	26.6	35.5
2-1 mm	%	40.8	55.5
0.5-0.25 mm	%	32.6	9.0

Table 1 Properties of the composts

Table 2 Resin	P, total P	and microbial	l P concentrat	tions of mulch	ned composts	on days (
and 75 (n =4)	. The LSD	refers to $P \leq$	0.001.			

Compost	Days	Resin P (mg kg ⁻¹)	Total P (mg kg ⁻¹)	Microbial P (mg kg ⁻¹)
C1	0	66.3	2100	59.1
	75	76.2	1800	48.1
C2	0	94.4	1000	84.3
	75	194.9	700	26.2
LSD (5%)		2.6	1.6	5.8

Table 3 Phosphorus concentration (mg kg⁻¹) in different P pools in the unamended soil on days 0 and 75 (n=4). Values in same row followed by different letters are significantly different ($P \le 0.001$).

P pools	Day 0	Day 75
Water-soluble P	4.3 a	5.1 a
Microbial P	4.6 a	5.9 a
NaHCO ₃ -Pi	10.8 b	6.8 a
NaHCO ₃ -Po	3.1 a	5.2 a
NaOH-Pi	26.8 a	30.7 b
NaOH-Po	56.6 b	38.9 a
HCl-P	30.9 a	32.9 a
Residual P	53.9 a	63.1 b



Fig 1 Cumulative respiration in unamended soil (C) and soils amended with mulched (m) or incorporated (i) composts (C1-C2), rock phosphate (RP) and composts with rock phosphate (n=4) after 14 days in soils sampled on day 0 and 75. Lines at top of the bars indicate standard deviation. Thick vertical line indicates least significant difference (LSD)



Fig. 2 Shoot (A) and root (B) dry weight (g kg⁻¹) and shoot P concentration (g kg⁻¹) (C) and P uptake (g pot⁻¹) (D) of wheat grown for 75 days in unamended soil (C) and soils amended with composts mulched (m) or incorporated (i) (C1-C2), rock P (RP) and composts with rock P (n=4).. Lines at top of the bars indicate standard deviation. Thick vertical lines indicate least significant difference (LSD).



Fig 3 Concentration of P pools in soils amended with composts (C1-C2), rock P (RP) and rock P with composts as mulch (m) and incorporation (i) in the soil on days 0 and 75 after subtracting the values of the unamended soil (n=4).

Chapter 5

Compost effects on microbial activity and biomass and soil P pools as affected by

particle size and soil properties

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The work contained in this chapter has been accepted in Journal of Soil Science and Plant Nutrition

STATEMENT OF AUTHORSHIP

Compost effects on microbial activity and biomass and soil P pools as affected by particle size and soil properties

Suman Lata Verma (Candidate)

Performed experiment, analyses of soil and composts, data analysis and interpretation, wrote manuscript, acted as a corresponding author.

I hereby certify that the statement of contribution is accurate.

Signed:

Date: 22 04 2.013.

Petra Marschner

Supervised development of work, data interpretation and manuscript evaluation and correction

I hereby certify that the statement of contribution is accurate and I give permission for the inclusion of the paper in the thesis.

Signed:

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Compost effects on microbial biomass and soil P pools as affected by particle size and soil properties.

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Abstract

Previous studies have shown that finer composts release nutrients more quickly than coarse composts, but little is known about the nutrient release from different size fractions of a compost. To assess if compost size fractions differ in their effect on microbial biomass and soil P pools, a compost from garden waste was separated by sieving into different particle size fractions (> 5 mm, 3-5 mm and < 3 mm) and then mixed into three different soils (referred as soil A, B and C) at a rate of 50 mg total P kg⁻¹. These soils were chosen because the size of soil P pools could be affected by soil properties which may also influence how the P pools respond to compost addition. Cumulative soil respiration, microbial biomass and soil P pools were determined after 25 and 50 days. Compost addition increased soil respiration and micobial biomass C and P, with the greatest increase in respiration with the > 5 mm fraction and in microbial biomass C and P with the < 3 mm fraction. The size of the P pools was mainly affected by soil type, the compost fractions had similar effects on soil P pools except for a greater increase in available P concentration by the finest fraction compared to the coarser size fractions. Soil A with the highest organic matter content had the highest concentrations of Fe/Al associated P (NaOH-P) and organic P whereas Ca-associated P (HCl-P) dominated in the alkaline soil C. The size of most P pools, even the so-called stable P pools changed over time in amended and non-amended

soils. It can be concluded that the size of P pools is more strongly affected by soil properties such as organic matter content, texture and pH than by the compost fractions.

Key words: size fractions, soil properties, temporal changes

1. Introduction

Phosphorus deficiency is a major constraint for crop production in many soils due to the poor solubility of P salts and P fixation which lead to low P availability even when total P concentrations in the soil are high. Fertilisers with high concentrations of soluble P are used to increase plant P availability, but P availability rapidly decreases as fertiliser P reacts with Al and Fe in low pH soils and with Ca in high pH soils (Bertrand et al., 2003; Khan and Joergensen, 2009). This insoluble accumulated P can be released in particulate form into surface water and lead to eutrophication (Song et al., 2007).

Application of P in the form of organic materials such as manures, plant residues and compost may provide alternative P sources for plants directly and indirectly. Directly by adding P to the soil and indirectly by release of organic acids, which block P adsorbing sites in the soil, changes in soil pH and complexation of soluble Al and Fe (Ayaga et al., 2006; Hue, 1991). The effect of added organic material on soil P availability not only depends on its P concentration but also on soil type, solubilty and mineralisation of organic P, C/P ratio (Peters et al., 2011) but also on particle size (Gómez-Muñoz et al., 2011). Large particles have a smaller surface area to volume ratio and are therefore less accessible to microbes than the finer particles. Additionally, the coarser fractions may have lower N and P concentrations than the finer fraction because of the pre-dominance of C-rich compounds (Tognetti et al., 2008). In a previous study it has been shown that finer composts released more N and P than coarse compost (Duong et al., 2012). Compost application can also indirectly

increase nutrient availability by increasing microbial activity and abundance (Pascual et al., 1997; Annabi et al., 2007) and enzyme activity (Ferreras et al., 2006) which can be explained by the supply of labile organic C with compost (Annabi et al., 2007) and the increased total soil organic carbon content (Pascual et al., 1997; Ferreras et al., 2006). The relationship between particle size of composted organic material and C and N dynamics in the soil was studied by Doublet et al. (2010) and Fangueiro et al. (2008) and N mineralisation was greater in fine particles of manures and compost (Tester et al., 1979). However little is known about the effect of different compost fractions on P release and P pools of soils differing in properties. The comparison of different soils affect compost decomposition and the size of P pools. The aim of the present study was to determine the effect of different size fractions derived from one compost on microbial biomass and P pools in soils with different texture, organic matter content and pH.

2. Materials and methods

2.1 Experimental setup

The three different soils used in this experiment were collected from 0-10 cm at Mount Bold, South Australia ($35^{\circ}06'53''$ S and $138^{\circ}41'18''$ E) and Birchip, Victoria (between latitudes $35^{\circ}10'$ and $39^{\circ}54'$ S and longitudes $143^{\circ}04'$ and $02^{\circ}78'$ E) (Table 1). The soils were air dried and sieved to < 2 mm. Before the start of the experiment, the soils were pre-incubated for 10 days at 55% water holding capacity to reactivate the microbes. This water content was based on Setia et al. (2011) who determined the water content that resulted in maximal soil respiration for soils of different texture and found that for soils with 10-20% clay 55% water holding capacity resulted in the highest respiration. Compost from garden waste was separated by dry sieving into the following fractions: > 5 mm, 3-5 mm and < 3 mm. These fractions were air-dried and analysed for various properties (Table 2). The different compost fractions were added to the soils at a rate of 50 mg total P kg⁻¹ soil and throughly mixed into the soils. The controls (without compost addition) were mixed similarly. The soil (30 g) was filled into PVC cores (radius 1.85 cm and height 5 cm) with a nylon mesh base (0.75 μ m. Australian Filter Specialist) and incubated at 22-25°C for 50 days. On days 0, 25 and 50, destructive samples were taken to measure microbial biomasss C and P pools. The samples at day 0 were taken approximately 5 h after compost addition. At each sampling date, there were four replicates per compost fraction and soil.

2.2 Soil and compost fraction analyses

Soil particle size was determined by the hydrometer method (Ashworth et al., 2001). Electrical conductivity and pH were measured in a 1:5 soil water ratio after 1 hour endover-end shaking at 25°C. Total organic C was analysed with potassium dichromate and sulphuric acid digestion method as described by Walkley and Black (1934). The water holding capacity of the soils was measured using a sintered glass funnel connected to a 100 cm water column (ψ m = -10 kPa) (Klute, 1986).

Soil respiration was measured with a Servomex 1450 series food pack infrared gas analyser from day 0 to 25 for cores to be destructively sampled on day 25 and from day 26 to day 50 for cores to be destructively sampled on day 50. On day 0, the cores to be sampled on day 25 were immediately placed in 1L glass jars closed with air tight lids equipped with septa to measure respiration. The remaining cores were incubated under

the same temperature conditions in large plastic containers. On day 25 the cores placed in jars on day 0 were removed and replaced by those which had been incubating in the plastic containers. The second set of cores was destructively sampled on day 50. The water content of the soils was maintained by weight, water loss from the soils was minimized by placing containers with water in the jars and large plastic containers. For determination of soil respiration, the CO_2 concentration in the headspace was measured after sealing the jars (t0). The closed jars were then incubated in the dark at 22-25°C for 1-4 days (shorter period at the start of the experiment, longer towards the end when respiration rates were low) and then second measurement of the CO₂ concentration was taken (t1). After the second measurement, the jars were opened to refresh the air using a fan. The CO₂ evolved from each sample was calculated as the difference between t1 and t0 for each measurement interval. The relationship between detector response and CO₂ concentration was determined by injecting known amounts of CO₂ injected into jars similar to those used for soil incubation and calculating the linear regression between detector reading and CO₂ concentration. The calculated CO₂ concentration was multiplied by the gas volume of the jars to obtain the mL of CO_2 -C respired during each measurement period and divided by the soil dry weight.

Microbial biomass C was estimated by fumigation-extraction (Vance et al., 1987) on days 25 and 50 after compost addition using 10 g soil. The soil samples were fumigated for 24 hr with ethanol-free CHCl₃ and extracted with 0.5M K₂SO₄. The C concentration in the filtered extracts was determined by titration with 0.0667M K₂Cr₂O₇ and sulphuric acid (Anderson and Ingram, 1993). The remaining dichromate was titrated with 0.033M acidified (NH₄)₂Fe(SO₄)₂·6H₂O. Chloroform-labile C was calculated as difference between fumigated and non-fumigated soil. To obtain microbial

biomass C, the chloroform-labile C concentration was multiplied by 2.64 (Vance et al., 1987).

Total N was determined by wet oxidation with sulphuric acid and hydrogen peroxide (Parkinson and Allen, 1975). Total P was measured after digestion with a mixture of nitric acid and perchloric acid (6:1) (Kuo, 1996) and P in digest was measured colorimetrically (Murphy and Riley, 1962). Available and microbial P were analysed using anion exchange resin as described by Kouno et al. (1995). Microbial P was calculated as the difference between the P concentration with or without hexanol (Kouno et al., 1995). Soil P pools were analysed using sequential fractionation as described by Hedley et al. (1982) and Tiessen and Moir (1993) with some modifications. One g of soil was placed in 50 mL centrifuge tubes and sequentially extracted (at 1:30 soil: solution ratio) with deionised water, 0.5 M NaHCO₃, 0.1 M NaOH, 1 M HCl and again 0.1 M NaOH. The two NaOH extracts were combined. The soil was shaken with the extractants on a reciprocating shaker at 200 rev min⁻¹ for 16 h, centrifuged (1610 \times g) and filtered through Whatman filter paper #42. Ten mL of filtered extract from the NaHCO₃ and NaOH fractions was acidified to pH 1.5 to precipitate organic matter. The supernatant was centrifuged (1610 \times g), filtered and analysed for inorganic P. Total P in these fractions was determined as described by Huang and Zhang (2009). Five mL of extract was digested with 1mL of 5 % potassium persulphate at 90°C for 16 hours which converts organic P into inorganic P and analysed for total P. Organic P of these fractions was calculated as difference between total P and inorganic P (measured in the undigested aliquot). Residual P from residual soil was determined by digestion with HNO₃-HClO₄ (6:1). The P concentration in the fractions was determined colorimetrically at 712 nm (Murphy and Riley, 1962).

2.3 Statistical analyses

The experiment was arranged in a complete randomized block design with three fixed factors (soil type, compost particle size and sampling time) and four replicates per treatment. The data was analysed by three-way analysis of variance (GenStat for windows 11.0, VSN Int. Ltd, UK, 2005). The LSD values for the interaction between type, compost particle size and time refer to $P \le 5$ %. Tukey test and linear regression were used to determine significant differences and correlations.

Principal component analysis was carried out using the P pools as variables and soil properties as environmental factors for each sampling date separately (Primer-E Ltd, Plymouth Marine Laboratory, Plymouth, UK).

3. Results

The soils differed in range of properties (Table 1). The Mount Bold soil (soil A) had the finest texture and lowest pH, but highest organic matter content and total N concentration. The two soils from Birchip (soil B and C) had similar sand content which was higher than that in soil A, but differed in pH. Soil B had a near neutral pH whereas soil C was alkaline. Compared to soil B, soil C had a higher EC and higher total P, total N and organic C concentrations.

The compost size fractions did not differ in pH and EC, but the available P concentration was highest in 3-5 mm fraction whereas the total P concentration was highest in the < 3 mm fraction (Table 2). However it should be noted that the total P concentration added was 50 mg total P kg⁻¹ soil for all three fractions. Total C and N

concentrations were highest in the < 3 mm fraction, but the C/N ratio was highest in the > 5 mm fraction. The amounts of C and N added were greatest in the < 3 mm fraction.

3.1 Soil respiration and microbial biomass

In the amended and unamended soils, cumulative respiration was highest in soil A and lowest in soil B (Fig. 1). Compost addition increased cumulative respiration in all soils with the increase being greatest with the > 5 mm fraction and smallest with the < 3 mm fraction. Compared to the unamended soil, the relative increase in cumulative respiration by compost addition was greatest in soil C. In soils B and C, cumulative respiration was greater from day 26 to day 50 than from day 0 to day 25.

On day 25, the microbial biomass C (MBC) concentration was similar in the unamended soils, but on day 50 it was highest in soil B (Fig. 2). Compared to the unamended soils, addition of all fractions increased the MBC concentration on day 25 in soil A whereas the MBC concentration was increased in soil B only the 3-5 mm fraction and in soil C by the 3-5 mm and > 5 mm fractions. On day 50, all compost fractions increased the MBC concentration compared to the unamended treatment in soil A whereas it was increased in soils B and C only some compost fractions: < 3 mm in soil B and > 5 mm and 3-5 mm in soil C.

3.2 Phosphorus pools

In the unamended soils, the available P concentration was lowest in soil A and highest in soil C (Table 3). Compost addition increased the available P concentration on days 25 and 50 with a significantly greater increase with the fine fractions (3-5 mm and < 3 mm) than the coarser fraction. In the amended soils, the available P concentration increased over time in soils B and C whereas it was highest on day 25 in soil A. The increase in available P concentration by the compost fractions on days 25 and 50 was greatest in soil A. Only in the unamended soil A, the available P concentration decreased over time.

In the unamended soils, the water-soluble P concentration was lowest in soil A and highest in soil B (Fig. 3). Irrespective of the size fraction, compost addition increased the water-soluble P concentration in soil A at all sampling dates. In soil B, addition of the 3-5 mm and < 3 mm fraction increased the water-soluble P concentration on day 25, but not on day 50. In soil C, addition of all fractions increased the water-soluble P concentration on day 25 whereas on day 50 it was increased only by the 3-5 and <3mm fractions. Only in soil A, microbial P increased over time. In this soil, addition of compost fractions increased the microbial P concentration on day 50. In soil B, the microbial P concentration on day 25 was increased compared to the unamended soil by the addition of the 3-5 mm and < 3 mm fractions and on day 50 by addition of > 5 mm and < 3 mm fractions. In soil C compared to the unamended soil, the microbial P concentration was increased only by addition of the 3-5 mm fraction on day 25.

The concentration of NaHCO₃-Pi was lower in soil A than in the other two soils whereas the reverse was true for NaHCO₃-Po (Fig. 3). Addition of the compost fractions increased the concentration of NaHCO₃-Pi on days 25 and 50 in all soils with greatest percentage increase in soil A. In soil A, the increase was not affected by compost particle size whereas in soils B and C, the increase was smallest with the coarsest fraction (> 5 mm). The concentration of NaHCO₃-Pi changed over time in the amended soils but differently in the three soils; with time it increased in soil A, decreased in soil B; and peaked on day 25 in soil C. Compared to the unamended soil, addition of compost fractions increased the concentration of NaHCO₃-Po on day 0 in soils B and C. On day 25, the NaHCO₃-Po concentration was increased in soil B by addition of the 3-5 mm and < 3 mm fractions and in soil C by the > 5 mm fraction. On day 50, only in soil A the addition of > 5 mm and 3-5 mm fractions increased the concentration of NaHCO₃-Po. The NaHCO₃-Po decreased over time in the amended soils B and C.

The concentrations of NaOH-Pi and Po were greatest in soil A (Fig. 4). Compost addition had little effect on the NaOH-Po concentration but increased the NaOH-Pi concentration in soil A. Both NaOH-Pi and Po concentrations were maximal on day 25 in the amended soil A, whereas there was little temporal change in soils B and C. On day 25 in soil A, addition of the 3-5 mm and < 3 mm fractions significantly increased NaOH-Pi concentration compared to the > 5mm fraction. The compost fractions did not differ in their effect on the NaOH-Po concentration.

The concentration of HCl-P was highest in soil C and lowest in soil A (Fig. 4). Addition of the compost fractions increased the concentration of HCl-P on days 25 and 50 in all soils with the greatest increase by the 3-5 mm fraction on day 50 in soils B and C. In all amended soils, the HCl-P concentration was significantly higher on days 25 and 50 than on day 0.

The residual P concentration was higher in soil A than in soils B and C (Fig. 4). Compost addition did not change the concentration of residual P and it did not change over time.

The PCA plots based on the P pools show a clear separation of soil A from soils B and C at all sampling dates (Fig. 5). This separation is based on the higher concentration of organic NaHCO₃ and NaOH, NaOH-Pi, total and residual P in soil A whereas the concentration of NaHCO₃-Pi, available P and HCl-P were higher in soils B and C. The separation of soils B and C along PC2 is based on higher concentrations of water-soluble P in soil B. The concentration of microbial P was higher in soil C on day 0 and 25, but on day 50 it was highest in soil A. The comparison of the loadings of the P pools with those of the soil properties (Table 4) shows that water-soluble P was positively correlated with sand content whereas the concentration of NaHCO₃-Po, NaOH-Po, residual and total P were correlated with silt and clay content as well as TOC and TN concentrations. The pH was correlated with the concentration of HCl-P (Table 4).

4. Discussion

The differences between the soils in the measured properties were greater than among the compost fractions. Compost addition increased microbial activity and biomass as well as the concentration of most P pools except NaOH-Po and residual P with only small differences among the compost size fractions. This study also showed that the P pools concentration is predominantly affected by soil properties such as organic matter, texture and pH.

Soil A differed in the concentration of the organic pools and total P from the other two soils and the PCA showed that this can be explained by its higher organic matter content, lower pH and fine texture. The high organic matter content also resulted in higher microbial biomass, cumulative respiration in soil A compared to the other two soils.

The higher pH in soil C can explain the higher concentration of HCl-P which represents P associated with calcium (Lee et al., 2004). The effect of soil texture on soil P pools was evident in the higher concentrations of water-soluble P in the two coarser textured soils compared to soil A. Sand would have fewer potential P binding sites than clay and silt, thus P would remain in water-soluble form in soils with a higher percentage sand. The fine texture of soil A is likely to have indirectly affected the size of the P pools by the higher organic matter content. The higher organic matter content can be explained by the increased plant input due to the higher soil fertility as well as protection of organic matter in aggregates (Baldock 2007). The higher proportion of NaOH-P in this soil which represents P associated with Al and Fe (Sikora and Corey, 1976) can be explained by the presence of clay which is usually coated with Fe and Al hydroxides (Baldock 2007).

4.1 Differences among compost fractions

The differences in the measured parameters among the compost fractions were small compared to the differences among the soils. Cumulative respiration was increased in compost amended soils compared to the control soils because of the organic matter addition with compost (Ros et al., 2006). The coarse compost fraction (> 5 mm) increased microbial activity more than the other fractions, but not microbial biomass. The higher activity occurred although less total C was added with this fraction, but because the C/N ratio of this fraction was higher, the additional C could not be used for build-up of microbial biomass and was instead respired. The large increase in microbial biomass in the first 25 days with finest fractions in soils A and C can be explained by the larger surface area to volume ratio of the fine particles which increases accessibility

to soil microbes (Magid et al., 2010). However this great accessibility did not result in higher cumulative respiration suggesting that the C was preferentially used to for growth rather than respiration. The larger surface area to volume ratio and higher decomposability of the fine fraction can also explain why the finest fraction induced the greatest increase in NaHCO₃-Pi and resin P concentrations in soils A and B. This suggests that compost P was mineralised to a greater extent during incubation from this fraction than from the coarser fraction. The difference between the compost fractions in NaHCO₃-Pi and resin P concentration with organic amendments was also reported by Schefe et al. (2008). In addition to P release during mineralisation this can also be explained by chelation of Al and Fe with organic acids formed during decomposition of organic compounds (Hu et al., 2005) which block P adsorption sites and may mobilise P from poorly soluble forms (Sanchez and Beck, 1994).

4.2 Changes in P pools over time

The concentration of some P pools that are considered as stable (NaOH-P, HCl-P) changed considerably over the course of the experiment suggesting that they are quite labile. Vu et al. (2010) also found that these stable P pools are converted into plant available P and Mat Hassan et al.(2012) found large changes in concentration of these pools in the rhizosphere of legumes and wheat. In the present study, only the residual P concentration did not change over time. The changes over time in the size of most P pools can be explained by P release from the added compost (in the amended soils) but

also by P flux among the pools, e.g., NaOH-Pi being converted to HCl-P and NaOH-Pi and NaHCO₃-Pi taken up by the microbial biomass (Ochwoh et al., 2005).

5. Conclusion

The study showed that the finest compost size fraction increased microbial biomass and P availability more than the coarser fraction which can be explained by its high surface area volume ratio and higher decomposability compared to the coarser fraction. However, the differences among compost size fractions with respect to their effect on soil P pools were small; the size of the P pools was mainly a function of soil properties such as organic matter content, texture and pH. It should be noted that we could only measure the net effect of compost addition on soil P pools. It would be of interest to follow P from organic amendments (e.g. plant residues) into soil P pools by using ³²P labelled plant residues.

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Properties	Unit	Mount Bold soil	Birchip	Birchip soil
		(A)	soil(B)	(C)
Sand	%	53	84.5	84.5
Clay	%	20	13.0	13.0
Silt	%	27	2.5	2.5
pH		$4.8^{\pm0.02}$	$6.1^{\pm 0.04}$	$8.1^{\pm0.01}$
Electrical	$dS m^{-1}$	$51.8^{\pm0.01}$	$50.2^{\pm0.20}$	$129.1^{\pm 1.14}$
conductivity				
Total organic C	g kg ⁻¹	$37.6^{\pm0.32}$	$5.5^{\pm0.06}$	$8.3^{\pm 0.13}$
Available P	mg kg ⁻	$11.0^{\pm0.07}$	$13.6^{\pm 0.33}$	$14.8^{\pm 0.24}$
	1			
Total P	g kg ⁻¹	$0.4^{\pm0.01}$	$0.1^{\pm 0.00}$	$0.2^{\pm 0.01}$
Total N	g kg ⁻¹	$2.9^{\pm0.07}$	$0.4^{\pm 0.01}$	$0.7^{\pm0.02}$
Bulk density	g cm ⁻³	1.4	1.6	1.6

Table 1 Properties of the soils used in the experiment (n=4, \pm standard deviation).

Properties	Unit	> 5 mm	3-5 mm	<3 mm
рН		$8.8^{\pm 0.01}$	$8.5^{\pm 0.01}$	$8.5^{\pm0.01}$
Electrical conductivity	dS m ⁻¹	$1.8^{\pm0.1}$	$1.9^{\pm 0.01}$	$1.9^{\pm0.01}$
Available P	mg kg ⁻¹	$26.9^{\pm0.9}$	$52.7^{\pm1.42}$	$44.9^{\pm 0.71}$
Available P added	mg kg ⁻¹	0.7	1.4	1.0
Total P	g kg ⁻¹	$1.8^{\pm 0.3}$	$1.8^{\pm 0.21}$	$2.3^{\pm0.09}$
Total P added	mg kg ⁻¹	50	50	50
Total N	g kg ⁻¹	$9.3^{\pm0.7}$	$12.8^{\pm0.95}$	$13.1^{\pm0.51}$
Total N added	g kg ⁻¹	0.3	0.4	0.3
Total organic C	g kg ⁻¹	$147^{\pm1.5}$	$149^{\pm0.71}$	$155^{\pm0.19}$
Total Organic C added	g kg ⁻¹	4.1	4.0	3.4
C/P ratio		82	83	67
C/N ratio		16	12	12

Table 2 Properties of compost fractions with particles > 5 mm, 3-5 mm or < 3 mm (n=4, \pm standard deviation)

Table 3 Available P concentration (mg kg⁻¹) in soils A, B and C amended with different compost particle sizes: > 5mm, 3-5 mm and < 3 mm and unamended soils (0) (n=4, \pm standard deviation). The LSD refers to the interaction of soil type, compost fraction and time.

Soils	Compost fractions	Day 0	Day 25	Day 50
A	> 5 mm	$11.9^{\pm 0.2}$	$16.7^{\pm 0.2}$	$10.6^{\pm 0.5}$
	3-5 mm	$12.9^{\pm0.1}$	$22.6^{\pm0.4}$	$17.4^{\pm 0.7}$
	< 3 mm	$12.6^{\pm 0.3}$	$23.2^{\pm0.2}$	$16.1^{\pm 0.1}$
	0	$11.0^{\pm0.1}$	$9.7^{\pm0.4}$	$6.2^{\pm0.1}$
В	> 5 mm	$14.2^{\pm 0.2}$	$15.7^{\pm 0.2}$	$17.1^{\pm 0.4}$
	3-5 mm	$14.6^{\pm 0.2}$	$19.1^{\pm 0.6}$	$22.6^{\pm0.5}$
	< 3 mm	$14.2^{\pm 0.2}$	$18.8^{\pm0.6}$	$18.3^{\pm0.4}$
	0	$13.7^{\pm0.3}$	$13.4^{\pm0.3}$	$13.4^{\pm0.2}$
С	> 5 mm	$15.5^{\pm 0.3}$	$17.3^{\pm 0.4}$	$17.4^{\pm 0.1}$
	3-5 mm	$16.1^{\pm 0.4}$	$17.8^{\pm 0.9}$	$22.2^{\pm0.6}$
	< 3 mm	$15.7^{\pm0.2}$	$18.5^{\pm0.6}$	$20.2^{\pm0.5}$
	0	$14.8^{\pm 0.2}$	$17.2^{\pm 0.1}$	$16.0^{\pm0.1}$
LSD			1.06	
Table 4 Loading of P pools and environmental factors for PC1 and PC2. For a given pool/factor only the loading of the axis with the higher loading value is shown. The loadings for the P pools are presented separately for each sampling date, the loadings for the environmental factors apply to all 3 sampling dates.

	Day 0 Day 25		y 25	Day 50		Environmental		
Variable	PC1	PC2	PC1	PC2	PC1	PC2	PC1	PC2
Water-soluble P	0.35		0.37		0.35			
Resin P	0.30			-0.60		-0.49		
Microbial P		-0.72		-0.55	-0.31			
NaHCO3-Pi	0.35		0.33		-0.35	-0.44		
NaHCP3-Po	-0.35		-0.37		-0.35			
NaOH-Pi	-0.35		-0.37		-0.34			
NaOH-Po	-0.34		-0.36		-0.33			
HCl-P		-0.53		-0.39		-0.55		
Residual P	-0.35		-0.37		-0.35			
Total P	-0.34		-0.37		-0.33			
Sand							0.99	
Silt							-0.99	
Clay							-0.99	
TOC							-0.99	
Total P							-0.94	
Total N							-0.98	
pH							0.79	
EC								-0.60



Fig. 1 Cumulative respiration (mg g⁻¹ soil) in soils A, B and C amended with different compost fractions: > 5mm, 3-5 mm and < 3 mm and unamended soils (0) from day 0 to day 25 and from day 26 to day 50 (n=4, the error bar on the columns is the standard deviation). Thick vertical line indicates LSD (interaction between soil type × compost fractions × days).



Fig. 2 Microbial biomass C (mg kg⁻¹ soil) in soils A, B and C amended with different compost fractions: > 5mm, 3-5 mm and < 3 mm and unamended soils (0) on day 25 and day 50 (n=4, the error bar on the columns is the standard deviation). Thick vertical line indicates LSD (interaction between soil type × compost fractions × days).



Fig. 3 The concentrations of microbial P, water-soulble P and NaHCO₃-Pi and Po (mg kg⁻¹) in soils A, B and C amended with different compost fractions : > 5mm, 3-5 mm and < 3 mm and unamended soils (0) on days 0, 25 and 50 (n=4, the error bar on the columns is the standard deviation). Thick vertical line indicates LSD (interaction between soil type × compost fractions × days).



Fig. 4 The concentrations of NaOH-Pi and Po, HCl-P and residual P (mg kg⁻¹) in soils A, B and C amended with different compost particle sizes: > 5mm, 3-5 mm and < 3 mm and unamended soils (0) on days 0, 25 and 50 (n=4). Thick vertical line indicates LSD (interaction between soil type × compost fractions × days).



Fig. 5 Principal component plots based on P pools on days 0, 25 and 50 showing the P pools as vectors (a, b and c) and the corresponding soil properties (d). The plot with the soil properties was the same for all three sampling dates. Symbols A, B and C refer to the three soils, the numbers next to the symbols indicate the compost treatment: 0 unamended control, 1: >5 mm fraction, 2: 3-5 mm fraction, 3: <3 mm fraction.

Chapter 6

Phosphorus uptake of wheat and size of P pools in four soils with or without

compost

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The work contained in this chapter is submitted in Pedosphere journal

STATEMENT OF AUTHORSHIP

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Performed experiment, analyses of soil, composts and plant samples, data analysis and interpretation, wrote manuscript, acted as a corresponding author.

I hereby certify that the statement of contribution is accurate.

Signed

Raj Setia

Contributed to planning of experiment, performed experiment and analysis of soil samples and plants.

I hereby certify that the statement of contribution is accurate and I give permission for the inclusion of the paper in the thesis.

Signed:

Petra Marschner

Supervised development of work, data interpretation and manuscript evaluation and correction

I hereby certify that the statement of contribution is accurate and I give permission for the inclusion of the paper in the thesis.

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Phosphorus uptake by wheat and size of P pools in four soils with or without compost

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Abstract

By taking up P from the soil, plants may affect the size of soil P pools and this effect may vary with soil properties and soil amendment. A short-term glasshouse experiment was conducted to compare P uptake of wheat and the size of soil P pools in four soils differing in pH, texture and organic matter content with or without compost. Four soils from two sites were used, with a heavier and a lighter textured soil from each site which were left unamended or amended with compost (from garden waste) as a 2.5 cm thick layer on the soil surface. Wheat was grown for 63 days. The soils unamended were sampled on day 0 and amended and non-amended soils on day 63. In all treatments, the concentrations of most P pools were higher in the two heavier textured soils (16 and 35% clay) than in the two lighter textured soils (8 and 13% clay). Compost addition increased shoot growth and P uptake but had little effect on soil P pool concentrations. Compared to the unamended soil on day 0, the concentrations of microbial P, NaHCO₃-Pi, NaOH-Pi, HCl-P and residual P on day 63 were higher whereas the concentrations of the two organic pools (NaHCO₃-Po and NaOH-Po) were lower. This suggests mineralisation of organic P pools and formation of inorganic P as well as microbial P uptake. On day 63, presence of plants increased the concentrations of microbial P, NaHCO₃-Pi, NaOH-Pi and residual P compared to unplanted soil without compost. It can be concluded that the size of the P pools is predominately affected by soil texture.

Plants enhance the formation of less labile P pools such as HCl-P and residual P which could serve as plant P sources in the long term.

Key words: clay content, organic matter, phosphorus pools, soil texture

1. Introduction

Phosphorus plays many essential roles in transfer of energy, respiration, photosynthesis and formation of cellular structure (Raghothama 1999; Richardson et al. 2009). Most soils have high concentrations of total P, but the concentration of available P in the soil solution is very low. Due to low mobility and availability of P in the soil, inorganic fertilisers are used to provide sufficient P to crops. But inorganic fertiliser use efficiency is low; plant uptake is < 20% of applied P in the year of application (Bolland and Gilkes 1998; Damodar Reddy et al. 1999). This low fertiliser efficiency is due to the rapid formation of different less labile P forms which are poorly available to plants (Guo et al. 2000). Excess use of inorganic fertilisers results in accumulation of total P in soil and movement of particulate P into water bodies where it can cause eutrophication (Correll 1998; Smil 2000).

Soil P dynamics are mainly affected by a combination of different factors such as soil type, organic amendments, plants and environmental conditions (Oberson et al. 1996). The effect of plants on soil P availability is related root hair length, size and distribution of the root system, root exudates, soil moisture, changes in pH and interactions with microbes in the rhizosphere (Magid et al. 1996). The effect of plants on P availability may vary in soils with different properties.

Studies have shown that the response of soils to compost application varies with soil type. In sandy soils compost has been shown to increase N mineralisation

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(Castellanos and Pratt 1981; Egelkraut et al. 2000), water holding capacity, nutrient availability and soil aggregation in sandy soils (Kumar et al. 1985; Mylavarapu and Zinati 2009), whereas in clay soils compost improved aeration and hydraulic conductivity (Avnimelech et al. 1990; Aggelides and Londra 2000). These changes can also affect plant growth and P uptake and therefore the effect of plants on soil P pools. The aim of this experiment was to assess wheat P uptake and soil P pools in four soils differing in texture, organic matter content, pH and nutrient concentration and to determine if this effect is altered by the application of compost mulch.

2. Materials and methods

2.1 Experimental design

Four soils with different properties, hereafter referred to K1, K2, B1 and B2, were collected from 0-10 cm depth at Karoonda, South Australia (35°05′S 139°53′E/ 35.083°S 139.883°E) referred to K1 and K2 and Birchip, Victoria (between latitudes 35°10' and 39°54'S and longitudes 1 43°04' and 02°78'E) referred to B1 and B2 (Table 1). These soils were selected because they are from long-term rotation trials on P deficient soils and because at each site, there are two locations which differ in soil texture and were planted with the same crops. The soils used here had been cropped with wheat in the previous year and where collected before sowing of the next crop. The soils were air-dried at room temperature and sieved to < 5 mm sieve size. Before the start of the experiment, the soils were pre-incubated at 75% (soil K1), 55% (soils K2 and B1) and 50% (soil B2) of water holding capacity for two weeks and then filled in 1 kg pots. These water contents were chosen based on Setia et al. (2011) who showed that microbial activity is highest at these water contents in soils of this texture. Pregerminated wheat seeds (*Triticum aestivum* cv. Krichauff) were sown in pots with

compost (treatment CP) and without compost (treatment P). The compost derived from garden waste was applied 2.5 cm thick layer on the soil surface corresponding to 144 g moist compost per pot. Additionally, control pots were left unplanted and without compost. There were four replicates per treatment. The properties of the compost are as follows: pH 8.2; electrical conductivity 4.3 dS m⁻¹; total organic C 121 g kg⁻¹; available P 66 mg kg⁻¹; total P 2.2 g kg⁻¹; C/P ratio 77 and C/N ratio 17. The pots were watered regularly by weight to maintain the water content. Soil and plant samples were collected after 63 days. On day 63 in compost amended pots, the compost layer was removed. Shoots were cut above the soil surface and roots were removed from soils. The underlying soil was analysed for P pools.

2.2 Analysis of soils and compost

The soils were analysed on day 0 (unamended only) and day 63 (unamended without plants or with plants with or without compost). Soil particle size was determined by the hydrometer method (Ashworth et al. 2001). Soils and compost pH and EC were measured in a 1:5 soil: water suspension after 1 h end-over-end shaking at 25°C. Total organic C was measured using wet digestion with K₂Cr₂O₇ and H₂SO₄, followed by titration with acidified (NH4)2Fe(SO4)2·6H2O as described by Walkley and Black (1934). Water holding capacity was measured using a sintered glass funnel connected to a 100 cm water column (ψ m=-10 kPa), thoroughly wetted and allowed to drain for 48 h. Dry weight of soils was determined after oven drying at 105°C for 24 h (Klute 1986).

Available P and microbial P were extracted using anion exchange resin strips according to Kouno et al. (1995). Microbial P was calculated as difference between samples with or without hexanol (Kouno et al. 1995). Phosphorus was analysed colorimetrically as described by Murphy and Riley (1962). The concentration of different P pools was determined by the sequential fraction method based on Hedley et al. (1982) and Tiessen and Moir (1993) with some modification. One g of soil was placed in 50 mL centrifuge tubes and sequentially extracted with water, 0.5 M NaHCO₃, 0.1 M NaOH, 1 M HCl and again 0.1 M NaOH (at a 1:30 mL soil: solution ratio). The two NaOH extracts were combined. Five mL of extract from NaHCO₃ and NaOH fractions was acidified to pH 1.5 to precipitate organic matter, centrifuged and the supernatant was analysed for inorganic P. Total P in these fractions was analysed by digestion of 5 mL extract with 5% potassium persulphate (K₂S₂O₈) at 90°C for 16 h as described by Huang and Zhang (2009). Organic P was calculated as the difference between total P (after digestion) and inorganic P (undigested extract). Residual P was determined by digesting the residual soil with HNO₃ and HClO₄ (6:1 ratio) (Kuo 1996). The P concentration in the fractions was determined colorimetrically at 712 nm wavelength (Murphy and Riley 1962). Total N content was determined with a LECO C-144 C analyser as described by Merry and Spencer (1988).

2.2 Plant analysis

Shoots were cut at the soil surface and rinsed with water to remove adhering soil. The shoots were oven-dried at 65°C for 3 days and then ground. To determine shoot P concentration, 250 mg of dry shoots were digested with nitric acid and perchloric acid (6:1). The P concentration in the digest was measured using vanado-molybdate method (Hanson 1950).

2.3 Statistical analysis

The data on day 63 was statistically analysed by two-way analysis of variance (Genstat[®] for Windows 11.0, VSN Int. Ltd, UK, 2005) with soil type and treatments

(plants with or without compost and unamended soils) as fixed factors. For the plant data the fixed factors were soil type and plants with or without compost. The LSD refers the significant difference at $P \le 0.05$. Tukey test was used to determine significant differences between treatments.

3. Results

3.1 Soil properties

Texture and pH were similar in Karoonda soil 2 (K2) and Birchip soil 1 (B1) (Table 2). The clay content was highest in B2 and lowest in K1, whereas the sand content was highest in soil K1 and lowest in soil B2. The total organic C concentration was highest in B2 and lowest in K1.The available P concentration was highest in K2 and lowest in B1 whereas the total P concentration was highest in B2 and lowest in B1.The total N concentration was higher in K2 and B2 than in K1 and B1.

3.2 Phosphorus pools in the unamended soil on day 0

On day 0 in the unamended soils, the concentrations of most P pools (except water soluble P and NaHCO₃-Pi) were higher in soils K2 and B2 than in K1 and B1 (Table 2). The concentration of water-soluble P was highest in K2 and lowest in B2. Soils K2 and B2 had higher concentrations of microbial P, NaHCO₃-Pi, the two NaOH P pools, HC1 P, residual and total P than the other two soils. On the other hand, the concentration of NaHCO₃-Po was higher in the two soils from Karoonda (K1 and K2) than in the Birchip soils (B1 and B2).

3.3 Plant P uptake

Wheat shoot dry weight in the unamended soil was lower in soils K1 and B2 than the other two soils but with compost amendment dry weight did not differ among soils (Table 3). Shoot dry weight was significantly higher with compost than without in all soils. In the unamended soils, the shoot P concentration was lowest in soil B1 and highest in soil K1 (Table 3). Compost addition increased shoot P concentration only in soil B2 whereas it was decreased compared to the unamended control in soil K1. With compost, the shoot P concentration was lowest in soil B1 and highest in soil K2. Without compost, shoot P uptake was lowest in soils B1 and B2 and highest in soil K2. Compost addition significantly increased shoot P uptake in all soils which was highest in soil K2 and lowest in soil B1.

3.4 Phosphorus pools in soils on day 63

The concentrations of water-soluble P, NaOH-Pi and Po and HCl-P on day 63 differed little from those in the unamended soil on day 0 (Figure 1). On the other hand, the concentrations of microbial P, NaHCO₃-Pi and residual P increased whereas the concentration of NaOH-Po was lower on day 63 than in the unamended soil on day 0. Compared to day 0, on day 63, the presence of plants increased the concentration of water-soluble P in soils K2, B1 and B2 but only when not amended with compost. The microbial P concentration was higher in the planted than the unplanted soil (without compost) irrespective of compost addition. In soils K2, B1 and B2 the concentration of NaHCO₃-Pi on day 63 was higher in the planted than the unplanted soil. On the other hand, the NaHCO₃-Po concentration in soils K1, K2 and B2 was lower in the planted compared to the unplanted soil. The concentrations of both NaOH-P pools were higher in the planted soil without compost than the unplanted soil whereas for the planted soil with compost, this was true only for NaOH-Pi in soils K1, B1 and B2. The concentration

of HCl-P was not affected by the presence of plants except in soil K1 where it was higher in the planted than the unplanted soil. The presence of plants increased the concentration of residual P and in the planted soils K2 and B2 it was higher without than with compost.

4. Discussion

This study showed that the soil P pool concentrations are mainly affected by soil properties and are further modulated by the presence of plants whereas compost addition had little effect.

The soils differed in P pool concentration, with higher P concentrations in the two finer textured soils (K2 and B2) compared to the soils with lower clay content. The former also had a higher organic matter content which can explain their higher concentrations of organic P pools. Texture and organic matter content appear to be more important for the P pool concentration than pH because soils K2 and B1 had similar pH but soil K2 had a higher concentration of most P pools than soil B1. Even the concentration of HCl-P, which is considered to be Ca associated P, was higher in soil K2 than K1 although the pH of soil K2 was lower. However, the pH difference between the two soils was small, pH 5.6 in soil K2 and pH 6.2 in soil K1. Soil B2 had the highest pH 8.3 and consequently the highest HCl-P concentration.

The increase in microbial P concentration from day 0 to day 63 in all treatments can be explained by (i) decomposition of labile organic P as the concentration of NaHCO₃-Po decreased from day 0 to day 63, and,(ii) in the planted soils, with C supply through root exudates and root turnover. The additional C supply by root exudates and root turnover

(Helal and Sauerbeck 1986) explains the higher microbial P concentration in the planted soils compared to the unplanted soil on day 63. Some of the NaHCO₃-Po may also have been converted into NaHCO₃-Pi because the concentration of this pool increased from day 0 to day 63. In the unplanted soil without compost, the concentrations of NaOH-Pi and HCl-P did not change between day 0 and day 63, but the concentration of NaOH-Po decreased (except soil K1) which corresponded to an increase in residual P from day 0 to day 63 (except soil K1). This suggests mineralisation of organic P and conversion into stable inorganic P (Erich et al. 2002; Ochwoh et al. 2005).

With or without compost, the presence of plants increased the concentrations of NaHCO₃-Pi and NaOH Pi from day 0 to day 63; the concentration of these pools was also higher also compared to the unplanted soil on day 63. Plants without compost also increased the concentrations of NaOH-Po and residual P compared to the unplanted soil on day 63 whereas this was not the case for the plants with compost. This is an agreement with previous studies in which the concentration of soluble P and NaOH-P was increased in rhizosphere soils of various species (Hinsinger and Gilkes 1997; Kirk et al. 1999a) which suggests that plants stimulate the formation of stable inorganic and organic P pools possibly by increasing microbial activity (Hinsinger 2001). Thus, although the plants removed P from the soil via P uptake, there was no consistent decrease in concentrations of the P pools except for the concentration of NaHCO₃-Po in three of the four soils. The lack of depletion of most P pools is in contrast to previous studies which have shown many P pools including labile and non-labile P pools are depleted in the rhizosphere (Guo et al. 2000; Mat Hassan et al. 2012). This discrepancy may be due to the sampling method. In the present study, the soil from the whole pot was thoroughly mixed before taking the samples for P fractionation. In the experiments

by Guo et al.(2000) and Mat Hassan et al. (2012) rhizosphere soil was used. Thus we cannot rule out that there was depletion in some P pools in the rhizosphere in our study. Although the presence of plants affected the size of the P pools compared to the unplanted soil, the differences between the soils remained the same as in the unplanted soil. The increase in the concentration of some P pools compared to the unplanted soil on day 63 (microbial P, NaOH Po, residual P) was greatest in soil B2 which had the highest concentrations of these pools among the soils. This suggests that plants can enhance the formation of some P pools that already have high concentrations in absence of the plants. Soils with high organic matter content also have a large microbial biomass because organic matter content and microbial biomass are positively correlated (Garcia et al. 2000). However plant growth and P uptake were lowest whereas the organic C concentration was highest in soil B2 compared to the other soils. Thus plants stimulated the formation of stable P pools and microbial P uptake in this soil but this P does not appear to be available to the plants.

Compost addition to plants increased plant growth and P uptake, but had little effect on the P pool concentration even in soil B1 which had the lowest P pool concentrations. This suggests that the P added with the compost was readily taken up by the plants and was not converted into soil P pools by the end of the experiment. A transient conversion of compost P into soil P pools before being taken up by the plants can of course not be ruled out. The higher P concentration and P uptake in plants with compost also suggests that compost not only supply P to plants but also N through mineralisation (Bending and Turner 2009; Egelkraut et al. 2000).

Conclusion

The soils differed in P pool concentrations which can be explained by differences in texture and organic matter content. The presence of plants altered the concentrations of some P pools but there was no consistent decrease in P pool concentration despite P uptake by the plants. Indeed the plants seem to enhance the formation of stable P pools. Compost addition to the plants increased their growth but had little effect on P pool concentrations by the end of the experiment indicating that P supplied by the compost was primarily taken up by the plants. The enhanced formation of stable P pools in the presence of plants could be beneficial in the long-term because they may taken up by the following crop.

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Properties	Unit	Karoonda		Birchip	
		K1	K2	B1	B2
Sand	%	89.5	82.0	84.5	57.0
Silt	%	2.5	2.5	2.5	8.6
Clay	%	8.0	15.5	13.0	34.4
pН		6.2	5.6	5.6	8.3
Total organic C	g kg ⁻¹	4.8	12.4	5.7	16.6
Available P	mg kg ⁻¹	16.7	27.6	10.9	18.2
Total P	mg kg ⁻¹	99.4	170.6	94.5	276.6
Total N	g kg ⁻¹	0.5	1.1	0.6	1.2
Bulk density	g cm ⁻³	1.6	1.5	1.6	1.4

Table 1 Physical and chemical properties of the four soils from Karoonda (K1 and K2) and Birchip (B1 and B2).

Table 2 Concentration of different P pools (mg kg⁻¹ soil) in the unamended soils from Karoonda (K1 and K2) and Birchip (B1 and B2) on day 0 (n=4). Values in the same row followed by different letters are significantly different ($P \le 0.05$).

23.4 c		
23.10	13.9 b	11.8 a
5.6 b	1.5 a	4.6 b
13.0 c	4.8 a	15.6 d
10.7 b	5.8 a	7.1 a
33.4 c	10.8 a	35.2 c
71.5 c	33.5 a	76.6 c
23.0 b	8.6 a	70.0 c
10.6 b	8.9 ab	30.7 c
170.6 c	94.5 b	276.6 d
	10.7 b 33.4 c 71.5 c 23.0 b 10.6 b 170.6 c	10.7 b 5.8 a 33.4 c 10.8 a 71.5 c 33.5 a 23.0 b 8.6 a 10.6 b 8.9 ab 170.6 c 94.5 b

Table 3 Shoot dry weight, P concentration and P uptake of wheat after 63 days growth in soils from Karoonda (K1 and K2) and Birchip (B1 and B2) amended with compost (n=4). Values in the same column followed by different letters are significantly different.

	Shoot dry weight (g pot ⁻¹)		Shoot P con	centration (mg kg ⁻¹)	Shoot P uptake (mg pot ⁻¹)		
Soils	Compost	No compost	Compost	No compost	Compost	No compost	
K1	1.27c	0.68a	1.67bc	2.25d	2.12cd	1.53b	
K2	1.21bc	1.02b	2.26d	2.13cd	2.73e	2.18cd	
B1	1.29c	0.99b	1.34ab	0.85a	1.71bc	0.84a	
B2	1.16bc	0.65a	1.95cd	1.24ab	2.26de	0.80a	



Fig. 1 Concentration of different P pools (mg P kg⁻¹): water-soluble P, microbial P, NaHCO₃-Pi, NaHCO₃-Po, NaOH-Pi, NaOH-Po, HCl-P and residual P on day 63 in soils from Karoonda (K1 and K2) and Birchip (B1 and B2) in unamended soil without plants (C) and with plants with compost (CP) or without compost (P) (n=4). Thick vertical bar indicates LSD for the soil × treatment interaction and thick horizontal lines indicate day 0 values.

Chapter 7

Conclusions and Future Research

Conclusions

Phosphorus (P) is an essential nutrient for crops but is poorly available in soils. To obtain adequate yield, farmers have to apply P fertilisers. Poor P fertiliser efficiency, eutrophication of surface water and depletion of high quality rock P reserves increased the interest in finding alternative P sources. The effects of organic P sources such as manure and plant residues on soil properties and plant P uptake have been studied (e.g. Fox et al. 1990; Iyamuremye et al. 1996; Erich et al. 2002; Ayaga et al. 2006), but less is known about the effect of compost, particularly with respect to soil P pools. Compost application has beneficial effects on soils and plants including improved water holding capacity and plant water availability (Giusquiani et al. 1995), increased soil structural stability (Annabi et al. 2007), soil N and P content (Epstein et al. 1976) and microbial activity (Ros et al. 2003). Understanding the effect of compost on soil P pools may help assessing the long-term effect on P availability because some P pools considered to be stable in the short-term can become available over time (Guo et al. 2000; Vu et al. 2008). Soils differ in P pool concentration (Tiessen et al. 1984); therefore the effect of compost on soil P pools may vary with soil type. It is known that co-composting of organic materials like manure and plant residues with rock phosphate can increase rock phosphate solubility (Zaharah and Bah 1997; Akande et al. 2005), but there is no information about effect of compost on rock phosphate when both are applied to the soil. Further, there is little information available on effect of different composts on different P pools and plant P uptake in soils and how this effect is modulated by soil properties. Therefore, the aim of the research described in this thesis was to determine

the effect of different composts on release of P pools with or without rock phosphate and P uptake by wheat.

In the first experiment (chapter 2), composts with different texture and nutrient concentration were applied as a layer on the surface to a loamy sand and their effects on soil respiration, soil P pools and plant P uptake studied after 14, 28 and 72 days. The results showed that compost mulch can act as slow release fertiliser supplying P and other nutrients, particular in the later growth stages. Composts with coarse texture and low available P concentration had a smaller effect on P pools and plant P uptake than the composts with fine texture and high available P concentration. Thus, application of composts with fine texture and high concentration of available and total P can increase soil P availability which could reduce the requirement of inorganic fertilisers for the crop to which the compost was applied and also for following crops.

To assess the effect of composts on solubility of rock phosphate, an experiment was conducted in loamy sand with two composts (from garden waste or from wood chips and bark) and rock phosphate for 75 days (Chapters 3 and 4). The composts were applied as mulch or incorporated into the soil to compare the effect of application method on solubility of rock phosphate. Rock phosphate was mixed into the soil. Application of composts with or without rock phosphate increased the concentrations of most measured P pools with greater effect with the garden waste compost which had a finer texture and higher total P concentration than the compost from wood chips and bark. Compost incorporation had a more pronounced effect on plant uptake and P pools than mulching. However, compost application did not increase rock P solubility. Since compost seem to supply adequate P to the plants and co-application with rock P did not alter the effect of the composts on soil P pools, there appears to be no benefit in applying rock P with compost.

To determine if effect of compost on soil microbial biomass and soil P is modulated by compost particle size and soil properties, an experiment was conducted for 50 days using three different soils and garden waste compost separated into three fractions (Chapter 4). This study showed that compared to the coarser fraction (> 5mm), the finest compost fraction (< 3 mm) increased microbial growth and P availability which can be explained by their higher high surface area to volume ratio and decomposability. The size of the soil P pools was mainly influenced by soil properties such as organic matter content, pH and texture. The soil with the high organic matter and clay content had higher concentrations of most P pools (except water-soluble P, NaHCO₃-Pi and HCl-P) than the two coarser textured soils. Compared to the soil properties, compost application had little effect on soil P pools and there was little difference between the size fractions.

In last the experiment, plants were grown in four soils with different properties with or without compost from garden waste and soil P pools and plant P uptake were studied after 63 days (Chapter 5). These soils had different properties compared to those used in the previous study and here the compost was not separated into different size fractions. The aim of this experiment was to assess the effect of plants on soil P pools in absence or presence of compost. This study showed that plants without compost changed the concentration of some P pools but despite plant P uptake, there was no consistent decrease in P pools concentration and plants increased the concentration of stable P pools (HCl-P and residual P). This indicates that plant influence the flux among P pools. Compost addition to plants increased growth and P uptake but had little effect on P pools suggesting that P supplied by the compost was taken up by plants. This experiment showed that in the soils used here, plants have a greater effect on P pools than compost. This is in contrast to the previous studies with plants (Chapters 2, 3 and

4) where compost application affected the size of the soil P pools compared to the unamended soil. Thus the compost effect on soil P pools is modulated by soil properties and also plants.

Future research

The experiments described in this thesis provide further insights in the effect of compost on soil P availability, soil P pools, plant growth and rock phosphate solubility. However, there are a number of points arising from this study that could be addressed in the future:

- The long-term effect of compost application on soil P pools and plant growth should be studied in field experiments where the compost is applied as mulch or incorporated once and soil P pools and plant growth studied over several growing periods. This will also show if compost has an effect after it is largely decomposed.
- To study the effect of mulch removal on soil P pools in absence of plants, an incubation experiment could be conducted where the compost is applied once, left for several weeks and then removed. Soil P pools could be measured at the start, just before removal of the mulch and at monthly intervals after mulch removal.
- Fluxes among P pools are likely to be affected by drying and rewetting cycles. Since compost affects both P pools and soil water content, it may also modulate the effect of drying and rewetting on soil P pools. Soil P pools and water content with or without compost exposed to three drying and rewetting cycles would be measured at the end of the drying phase and at the end of the rewetting phase.

The soils used in the experiments in this study had relatively low P fixing capacity. One of the mechanisms by which organic soil amendments increase P availability is blocking P fixing sites by organic acid anions. However, organic acid anions are mainly produced during organic matter decomposition. Compost is highly degraded and thus may release little organic acids compared to other organic amendments such as manure or plant residues. The effect of different organic amendments (compost, manure, plant residues) added at the same total P rate on P fixing capacity over time could be studied in soils with moderate to high P fixing capacity.

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