

Long-term compost application and soil P legacy impacts on enhancement of early maize growth

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Abstract

Purpose

Compost application is globally viewed as a long-term strategy to improve phosphorus (P) availability for agricultural crops. Limited information exists on the legacy effects of P in compost-amended soils concerning the subsequent effect of mineral P fertiliser on crop growth.

Methods

We therefore conducted two cycles of a maize growth experiments in pots (replicates; n=4) under greenhouse conditions each lasting 84 days. We used two soils that previously had been fertilized with compost over the past 10 years. The first cycle of maize growth was carried out

under full P fertilisation, i.e. 150 mg P kg⁻¹ soil (300 kg P ha⁻¹), as calcium dihydrogen phosphate, the second cycle of maize was grown in the same soil and pots after harvest of the first cycle. Plant and soil measurements did include plant P uptake soil Hedley P fractions.

Results

The easy plant available P NaHCO₃-P_i ranged from 93-221 mg P kg⁻¹, incrementing a 31% due compost and 76% due Ca-P fertilization against the control without compost and Ca-P fertilization, respectively. However, compost and fertilizer application increased dry matter production by only 7- 9% and plant P uptake by 11% and 17% compared to past compost application and Ca-P application, respectively.

Conclusion

Compost had a protective role against the depletion of NaHCO₃-P_o pool and thus was helpful to initiate a P legacy stock, thereby enhancing soil resilience and limiting P depletion by future high P demanding crop rotations. However, compost mixed with mineral P is the best P availability for both the immediate crop needs and longer term soil fertility preservation.

Keywords P fractionation • organic fertilizer • corn • water soluble calcium-P • legacy P

1 Introduction

Plant uptake of P and P use efficiency in agricultural systems are to a large extent limited by the availability of phosphate in the soil. Depending on soil characteristics, added P is mainly converted into recalcitrant forms due to reactions with aluminium, iron or calcium, sorption to and occlusion by oxides and binding in organic matter. The controlling processes of P availability in soil are (1) the adsorption/desorption of P, as organic and inorganic ligands compete with P for

the same adsorption sites onto Fe and Al oxides (e.g. Borggaard et al. 2005; Li et al. 2016), (2) the pH-dependent dissolution of P-containing primary minerals such as hydroxyapatite (e.g. Goyne et al. 2006) and secondary minerals such as Ca-, Fe- and Al-phosphates, and (3) enzymatic hydrolysis, which can be additionally involved in the above abiotic processes (e.g. Guppy et al. 2005). Although much research has been conducted during the last decades, our level of understanding processes concerning the release of P remains incomplete, while the complexity in the plant-soil system and the dependence of environmental conditions makes it difficult to forecast P release.

Sequential chemical extraction procedures, such as the Hedley soil P fractionation, separate soil P according to its attributed inorganic (P_i) and organic (P_o) soil fractions (Hedley et al. 1982; Cross and Schlesinger 1995). The sequential extraction is considered to follow a decreasing order of bioavailability (Hedley et al. 1982). The inorganic P-fraction extracted with 0.5 M sodium bicarbonate ($\text{NaHCO}_3\text{-}P_i$) is attributed usually to labile P_i in the solid soil phase consisting of P bound to Al and Fe surfaces (Cross and Schlesinger 1995). Moderately labile P_i extracted with 0.1M NaOH ($\text{NaOH-}P_i$) comprises chemisorbed P_i of lower bioavailability associated with amorphous and crystalline Fe- and Al-hydroxides (Hedley et al. 1982). Non-labile primary Ca-bound P_i is extractable through 0.5 M HCl or H_2SO_4 (Cross and Schlesinger, 1995). Regarding organic fractions, easily mineralizable (labile) $\text{NaHCO}_3\text{-}P_o$ and moderately labile $\text{NaOH-}P_o$ comprise P linked to organic matter (Cross and Schlesinger 1995). However, every fraction represents partly desorbed P from various soil constituents and partly P co-released upon dissolution of various minerals (Klotzbücher et al. 2019).

In organic and particularly in bio-dynamic farming, compost is regarded as a valuable source of P for crops. The addition of compost leads to an increase in total P as well as increased availability of P as humic acids and organic ligands in the compost material release inorganic P

(Dou et al. 2009; Oburger et al. 2009; Miller et al. 2019) through the ligand exchange of metal hydroxides such as Fe- and Al-phosphates that are exchanged against organic ligands (specific P-exchange) (Borggaard et al. 2005) as well as from Ca-phosphates by Ca-bounding (Singh and Amberger 1990). In this sense, effects of compost application can enhance soil macronutrients and P legacy in soil for further crops (Arruda- Coelho et al. 2019; Miller et al. 2019).

There are quiet few studies (e.g. Ojo et al. 2015; Song et al. 2017) that show the effect of compost application effect over soil P fractions distribution affecting plant growth. The 1st author found that compost incremented organic P and the 2nd author found that mineral fertilizer application incremented P_i fractions with enhanced growth with mineral fertilizer. Other studies only cover soil aspects, e.g. Bhat et al. (2017) shows an incremented soil biological activity under long term organic management, contributing to P availability.

Therefore, we hypothesize that a long-term application of compost changes the chemical properties of a soil, improving organic matter content, increasing pH and the distribution of P between the Hedley-fractions of soils, incrementing the amounts of P_o fractions (legacy effect), thus enhancing the early growth of maize (*Zea mays*). Consequently, the aim of this study was to examine how a long term past application of compost and soluble P fertilizer may change the distribution of P between the Hedley-fractions and influence the growth of maize.

2 Material and Methods

2.1 Former compost field experiment and soil sampling

We used soil from the Ap horizon (0-20 cm) of a Loess-derived Luvisol (48° 43' 00" N; 9° 11' 40" E) at the experimental farm "Heidfeldhof", belonging to the University of Hohenheim, in

Stuttgart, Germany (407 meters above sea level, 9.7°C average temperature, 736 mm average rainfall, “Cfb”- marine west coast climate Köppen climate classification subtype), derived from a past long term compost field experiment that lasted over 10 years. The former field experiment consisted in a Silage Maize (cv. Noveta)– Winter Wheat (cv. Asketis) – Spring Barley (cv. Scarlett) crop rotation with/without long term garden plant residues digested in a central compost plant. The compost application rate was approx. 40 t DM ha⁻¹ yr⁻¹ with 400 kg N ha⁻¹yr⁻¹. Compost added soils and no added soils differed distinctly in soil chemical properties, with higher pH, total C_o, P_T and CAL-P in the compost added soil (Table 1, for further soil characterization see Redel et al. 2013). From this experiment, we have taken samples from the different plots to make a composite sample from the compost added soil and no compost added soil, that we used for the present experiment.

2.2 Present 2-factorial pot-experiment

We conducted a 2-factorial pot-experiment (Factor 1: soil with, (C1) and without P fertilization (C0) respectively) x Factor 2: with/without application 150 mg P (kg soil)⁻¹ as Ca(H₂PO₄)₂x 2H₂O, (P1 and P0, respectively), applied as nutrient solution to the pots (4 kg soil pot⁻¹) before sowing. We performed a pot experiment (Mitscherlich pot, with 20 cm diameter and 20 cm height, 6 L capacity) with 4 replications under greenhouse conditions (day (16h): 20-25°, night (8h): 10-15°, 15 - 95% humidity, 130-180 W m⁻² light) with a water content maintained at 65% water holding capacity. Therefore, we have four treatments, namely C0P0 (no compost added soil and no P), C0P1 (no compost added soil with P), C1P0 (compost added soil and no P) and C1P1 (compost added soil with P).

In order to measure exhaustion of P fractions in soil induced by plant growth, two consecutive cycles of maize (cv. Aventina, 5 plants pot⁻¹, each for 84 days until booting) were cultivated with a base fertilization (70 mg kg⁻¹ 0.5M NH₄NO₃, 90 mg kg⁻¹ 0.25M K₂SO₄, 10 mg kg⁻¹ 0.25M MgSO₄•7H₂O) to avoid background nutrient effects.

2.3 Laboratory analysis and statistics

Plant material (shoot and root after washing) from the experiment was analysed for dry matter and total P concentrations (VdLUFA 1996) after the first and the second cycle of maize. At the beginning of the experiment (T0), soil was analysed for soil pH (0.01M CaCl₂), total carbon (C) and nitrogen (N) (Multi N/C 2100, Analytik Jena). Total organic C and N were determined with a C-N-auto-analyser (Elementar vario MAX CN). At T0, and after the first and second harvest of maize (M1; M2), total P (aqua regia-extracted (VdLUFA 2002)), P-Fractions (P extraction for 16 h with 0.5 M NaHCO₃, followed by 0.1 M NaOH and H₂SO₄ 0.5 M (Hedley et al. 1982; Tiessen and Moir 1993)) and CAL-P (VdLUFA 2002) were analysed. Inorganic P (P_i) in the soil extracts was determined photometrically according to Murphy and Riley (1962) and organic P was determined as the difference between total P measured by inductively coupled plasma optical emission spectrometry (ICP-OES–P) and P_i. We performed a 3-way ANOVA with previous compost application, fertilization and cycle as main factors. Kolgomorov test was applied for testing data normality. Differences between observation means were determined by Duncan's multiple range test ($\alpha = 0.05$).

3 Results

3.1 Plant parameters

During the experimental period, we determined a slight significant increment of above-ground biomass with compost application and P fertilization, with a calculated average of 85 g pot⁻¹ and 93 g pot⁻¹ for without (C0) and with compost (C1), respectively (Table 2 and 3). Significant differences were determined for the shoot concentration of P between P0 (calculated average of 1352 mg P kg DM⁻¹) and P1 (1731 mg P kg DM⁻¹) treatments and both factors, compost and phosphorus application affected significantly overall root P concentration means (Table 3). In general, plants showed indications of P deficiency in the first 3-4 weeks after emergence, especially those in the unfertilized treatment.

The overall differences in plant P uptake were less pronounced, with only an 11% increase associated with past compost application and 17 % associated with immediate Ca-P application (C0P0 < C1P0 < C0P1 = C1P1, Table 2 and 3). CAL-P was significantly enhanced both through the long-term application of compost (+87 mg P kg⁻¹) and P fertilizer (+73 mg P kg⁻¹) for the experiment. The successive growth of maize led to a decrease of CAL-P over time (-109 mg P kg⁻¹, Fig. 1).

3.2 Soil parameters

Long term compost application and P fertilizer application led to significant increases in pH and in the Hedley fractions NaHCO₃-P_i, NaHCO₃-P_o and NaOH-P_i. (Table 1 and Fig. 1). The NaOH-P_o fraction, however, decreased after compost application and did not change significantly after P fertilization (Table 3).

The addition of mineral P fertilizer led to an increase mainly in the easily plant available P $\text{NaHCO}_3\text{-P}_i$ fraction of the soil that ranged from 93-221 mg P kg^{-1} with $\text{C0P0} < \text{C1P0} < \text{C0P1} < \text{C1P1}$ (Table 2 and Fig. 1). This fraction was on average 31% higher in the compost soil compared with no compost treatment, and 76% higher in Ca-P fertilised soil (P1) compared with no P treatment (Table 3).

Moderately available plant P (NaOH-P_i Hedley fraction) was further increased after the first cycle of maize (M1) by a 23%. This fraction was on average 20% higher in P fertilized than in unfertilized treatment, and ranged from 144-177 mg P kg^{-1} NaOH-P_i ($\text{C0P0} = \text{C1P0} < \text{C0P1} = \text{C1P1}$, Table 2 and Fig. 1). After two growth cycles of maize, a depletion of the $\text{NaHCO}_3\text{-P}_i$ and NaOH-P_o fractions of 27% and 73% was observed (Table 3). This change was more enhanced in the second growth cycle compared to the first. Compost enhanced NaOH-P_o to P_i transformations (with averages of 42 mg kg^{-1} for C0P0 , 21 mg kg^{-1} for C1P0 , 32 mg kg^{-1} for C0P1 and 21 mg kg^{-1} for C1P1 , not shown). In contrast, an increase of the $\text{NaHCO}_3\text{-P}_o$ fraction by a 24 % was observed after M2 (Fig. 1).

Non-labile $\text{H}_2\text{SO}_4\text{-P}_i$ and $\text{H}_2\text{SO}_4\text{-P}_o$ was 248 mg kg^{-1} and 48 mg kg^{-1} in average for C0 , respectively, and 311 mg kg^{-1} and 64 mg kg^{-1} for C1 , without differences induced through fertilization and the crop cycle (Table 3).

3.3 P balance

The P balance was calculated as the difference between P added by fertilization and plant P uptake was between -52 mg kg^{-1} and +94 mg kg^{-1} , being positive for both fertilized treatments and negative for the both unfertilized ones (Table 2).

The relationship between plant P uptake, P fractions and soil parameters were determined before and after the respective cropping cycles (Table 4). We found that in general the first three fractions ($\text{NaHCO}_3\text{-P}_i$ and P_o and NaOH-P_i) sampled before the start of the crop cycle for correlated best with shoot P uptake. Overall, the highest correlations were between shoot P uptake and NaOH-P_i before cropping ($r = 0.74$, $P < 0.01$) and shoot P uptake and NaOH-P_o after cropping ($r = -0.88$, $P < 0.01$) (Fig. 2a and 2b). The P balance was positively correlated with $\text{NaHCO}_3\text{-P}_o$ ($r = 0.74$, $P < 0.01$), but negatively with $\text{NaHCO}_3\text{-P}_i$ and NaOH-P_o ($r \leq -0.71$, $P < 0.01$, Table 4). Finally, PCA analysis did show a close association between NaOH-P_i and P uptake and dry matter production for the PC2 axis. Whereas, the other P parameters (CAL, $\text{NaHCO}_3\text{-P}_i$ and P_o , and NaOH-P_o), were clustered together on the PC1 axis (Fig 2c).

4 Discussion

4.1 Long-term compost application effects on soil P fractions

In accordance with previous investigations (Lopez-Contreras et al. 2007; Malik et al. 2013), the legacy of past, long-term application of compost significantly affected the chemical properties of the soil. This is mainly with regards to inorganic P fractions, as added organic P is slowly mineralized over time (Malik et al. 2012). The enhancement of several P fractions due NaOH-P_o to P_i transformations was also found by Kashem et al. (2004), despite a higher organic matter content, found in our experiment (Table 1) but contrary of our hypothesis. This gradual turnover of organic P from compost led to an increase in CAL-extractable P, which is commonly used as an indicator for plant P availability in soils (Qayyum et al. 2015; von Tucher et al. 2018).

Compost added soil showed a higher acid phosphatase activity and microbial C and N (Redel et al. 2013), that together with a higher pH can support a greater microbial activity that favoured P absorption by the roots, in conditions that in overall the plants showed indications of P deficiency. Indeed, compost addition resulted in an increment of both available P concentrations (CAL-P and $\text{NaHCO}_3\text{-P}_i$ and P_o , Table 3). The available P increment can be attributed to P transformation processes derived from NaOH-P_o , that were found depleted with compost addition. This process is driven by improved soil biological activity, specifically higher enzymatic activity under organically managed soils as found by Bhat et al. (2017). Otherside, legacy-P derived from compost application incremented the most not available P fractions from 399 mg kg^{-1} to 505 mg kg^{-1} , with an 18 % increment of $\text{H}_2\text{SO}_4\text{-P}_i$ and P_o fraction (Table 3) or 19% of none labile P fractions (calculated from the total P- labile and moderate labile P fractions. In this sense, Dou et al. (2009) have found that this preservation in heavily manured soils occurs mainly as inositol hexaphosphate. Legacy P derived from long term compost application to the soil may limit P depletion by plant uptake and increase soil resilience against high P demanding crop rotations, thus enhancing none labile fractions that can be acting like a reservoir of P.

Finally, NaOH-P_i , which is tightly bound to Fe and Al oxides and clay minerals may slowly diffuse into the minerals and transform or precipitate in acid-P pools or can be transformed to more available P fractions (Joshi et al. 2018), did not change due compost addition (Table 3).

4.2 Mineral P fertilization effects on P fractions

Mineral P fertilization led to an immediate increase of CAL-P, as well as the $\text{NaHCO}_3\text{-P}_i$ and P_o fractions, as shown for T0 (Fig. 1), with almost all the added fertilizer was found in the increment of soluble and labile NaHCO_3 and NaOH-P_i fractions as the showed increment of P of COP1 and C1P1 in Fig. 3. In this sense, added mineral P is mainly determinable in the available fractions, as has been determined previously (Motavelli and Miles 2002; Muhammad et al. 2007). Fertilisation exerted similar effects in both compost treatments, increasing labile + moderately labile P_i (both with an increase of 148 and 141 mg kg^{-1} when we compare average means of P1 vs. P0, Fig. 3). However, P_o content after fertilization was reduced in C0 by 27 mg kg^{-1} , contrasting with the increment of 24 mg kg^{-1} with C1, (Fig. 3) thus reinforcing the protective role of compost against P depletion as stated above.

Regarding the non-labile fractions, $\text{H}_2\text{SO}_4\text{-P}_i + \text{P}_o$ incremented with fertilizer and compost application (Table 3). This result shows us that P was become fixed to the soil, even in conditions of P shortage to the plants. Even a part can become in the residual fraction, but we cannot calculi it, because we have not determined the total P in fertilized soils.

4.3 Effects of time and repeated maize cycle on P depletion

Mineral P incremented CAL-P and $\text{NaHCO}_3\text{-P}_i$ immediately to a maximum after the pot preparation and before sowing in all treatment. However, thereafter CAL-P declined to a minimum at M1, showing a fast P fixation to the soil, leading to an increment of the average concentration from NaOH-P_i from 161 mg kg^{-1} at M0 to 197 mg kg^{-1} at M1. Finally, NaOH-P_i dropped to an average of 122 mg kg^{-1} and NaOH dropped from 62 mg kg^{-1} at M0 to 16 mg kg^{-1} at M2 (Table 3). $\text{H}_2\text{SO}_4 \text{ P}_i + \text{P}_o$ fractions show us that P was slowly transformed to this fraction (average of 314 mg kg^{-1} for T0, 339 mg kg^{-1} for M1 and 354 mg kg^{-1} for M2, Table 3). This

result confirms us that P was become fixed to the soil, even in conditions of P shortage to the plants as we stated before. At M2, plants used until NaOH-P_o source, but were unable to use other fractions that were more recalcitrant. In this sense, we have found previously that compost incubation firstly improved labile and relatively labile fractions, but after 25 days P became fixed in non-extractable fractions (Redel et al. 2019).

4.4 P fractions as source for plant growth and its relation with P balance

The first three P fractions (NaHCO₃-P_i and P_o and NaOH-P_i) sampled at the start of the crop cycle, especially NaOH-P_i, exerted the most evident effect on plant P uptake (Table 4). However, NaOH-P_o concentration after the crop cycle was inversely correlated with P uptake, thus acting as a reservoir of P that depleted totally to 0 in some treatments (C0P0, C0P1 and C1P1) at the end of M2 cropping season. For this reason we did not draw NaOH-P_o in Fig 2b. Thus, NaOH-P_o uptake was stimulated by mineral fertilization as shown by the correlation of with P balance ($r = -0.69$, $P < 0.01$). Similar reductions of NaHCO₃-P_i and NaOH-P_i pools over a crop cycle were found by Cabeza et al., (2017) for an unfertilised early maize pot experiment. Additionally, Liao et al., (2020) also showed a depletion of these HCl-P_o and NaOH-P_i pools under P exhaustion conditions in long term maize field experiment. Furthermore, contrasting results in the response of P fractions to soil P depletions in maize have been attributed to a different weather and environmental conditions (Shi et al., 2015).

However, NaHCO₃-P_o was correlated positively with P input ($r = -0.71$, $P < 0.01$), showing that P fertilization improved labile P, probably from microbial biomass and organic acid production, which stimulated the breakdown of organic matter and simultaneously the conversion from NaOH-P_o to NaHCO₃-P_o fractions. Overall, as NaOH-P_i was not correlated with P inputs, it

shows that its uptake was soil but not fertilizer P. The importance of the NaOH-P_i fraction sampled before crop cycle in relation to P uptake and plant growth was also confirmed by the PCA analysis (Fig 2c). In this sense, NaOH-P_i comprises P that is held by Fe and Al and need to be solubilized through organic acids, and therefore is mainly depleted by crops as maize (Cabeza et al., 2017, Liao et al., 2020), and as we also find by wheat (Redel et al., 2007) and therefore its content at the beginning of the cycle is more important. NaOH-P_o is mainly P held by organic matter and can be stabilized by Al also (Redel et al., 2016), and mainly is accumulated in soil by microorganism activity denoting a sink of added P as we found for oat, barley and wheat (Redel et al., 2007). The depletion of NaOH-P_o in our experiment denotes a scarcity of soil P that is not common in other experiments.

4.5 Overall effects of fertilization on plant growth

We found that a 10% increase of aboveground-biomass formation in maize could be attributed to P derived from past (legacy) compost applications, mainly driven by an enhanced root P concentration as shown by Table 3. However, this increment was rather limited compared to the enrichment of P fractions in the soil, (e.g. NaHCO₃-P_i incremented a 31% due compost and 76% due Ca-P fertilization against the control without compost and Ca-P fertilization, respectively). Therefore, we observed a discrepancy between the assumed plant P availability based on Hedley extraction (NaHCO₃-P_i) and subsequent actual impact of P availability on plant growth, suggesting either that unfertilized plants were adapted to present conditions of soil P availability or that bio-chemical and physical characteristics of soils restrict the acquisition of P through maize, even when common soil chemical analysis suggests a high plant-availability of P.

Consequently, plant P absorption from soil was rather low, with plant tissue P concentration barely reached the threshold value of 0.8 g kg^{-1} proposed by Khamis et al. (1990) in unfertilized P treatments, but was two times higher in the +P treatments. However, Plénet et al. (2000) did also observed growth limitations with plant P concentrations over 2 g kg^{-1} . In contrast, Ahmad et al. (2013) observed that a similar P dose as in our experiment (135 mg kg^{-1}) was enough to reach a maximum in maize DM production with an average P concentration of 1.6 g kg^{-1} in plant tissue. Therefore, our research highlights the necessity to complement the indicators of plant P availability using Hedley P as indicator with soil biological and physical indicators.

5 Conclusions

Applied fertilizer stimulated NaOH-P_o conversion to $\text{NaHCO}_3\text{-P}_o$. However this was only partly taken up by plants and therefore accumulated in non-available P fractions, even under our experimental P scarcity conditions. Compost had a protective role against the depletion of $\text{NaHCO}_3\text{-P}_o$ pool and thus was helpful to initiate a P legacy stock, thereby enhancing soil resilience and limiting P depletion by future high P demanding crop rotations. However, compost mixed with mineral P is the best P availability for both the immediate crop needs and longer term soil fertility preservation.

Plant P uptake was mostly from NaOH-P_i , but its actual impact on maize growth was quite low. Therefore, our study results highlight a discrepancy between the assumed plant availability of P as per estimate by P extraction methods (Hedley P) and the actual impact of this perceived potentially available P on plant growth.

Funding Information This work was supported by the DAAD Research Stays for University Academics and Scientists, 2019 (57442043) and Becas Chile Bicentenario Grant from CONICYT (Comisión Nacional de Investigación Científica y Tecnológica de Chile). Dr. Hartmann is funded by the Deutsche Forschungsgemeinschaft (DFG, German Research Foundation) – 328017493/GRK 2366 (International Research Training Group "Adaptation of maize-based food-feed-energy systems to limited phosphate resources").

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

432 **Table 1** Effect of past compost application on soil chemical characteristics (means \pm standard error of 4
 433 analytical replications)

	pH	Total C_o	Total N_o	P_T	CAL-P
		---- g kg ⁻¹ ----		--- mg kg ⁻¹ ----	
C0	6.5 \pm 0.1	9.8 \pm 0.7	1.3 \pm 0.2	789 \pm 11	67 \pm 1
C1	7.2 \pm 0.1	17.7 \pm 1.3	2.1 \pm 0.3	922 \pm 13	144 \pm 4

434 Abbreviations: C0/C1: Silty loam soil without/with long-term compost application; Total C_o: total organic carbon; Total N_o: total
 435 organic N, P_T: total P; CAL-P: extractable with calcium acetate lactate solution.

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437 **Table 2** Analysed plant variables (Plant dry matter (g pot⁻¹), plant P concentration (mg kg⁻¹), plant P uptake (mg pot⁻¹) and P balance (mg kg⁻¹) of
438 maize in the 4 soil treatments at the 1st and 2nd harvest Different letters indicate significant differences between the treatments within the main factors
439 (α = 0.05, Duncan's multiple range test).

	Plant dry matter (g pot ⁻¹) ⁽¹⁾		Plant P concentr. (mg kg ⁻¹)		Plant P uptake (mg kg ⁻¹) ⁽²⁾		P balance (mg kg ⁻¹) ⁽³⁾		
	Shoot	Root	Shoot	Root	Shoot	Root	Input ⁽⁴⁾	Output ⁽⁵⁾	Balance
C0P0	81 ± 2 b	41 ± 3 a	1285 ± 60 c	1263 ± 14 b	26 ± 5 c	13 ± 4 b		39 ± 2 c	-39 ± 2 b
C0P1	88 ± 2 a	42 ± 2 a	1756 ± 37 a	1773 ± 100 a	39 ± 6 a	19 ± 3 a	150	58 ± 1 a	92 ± 1 a
C1P0	92 ± 3 a	47 ± 4 a	1419 ± 96 b	1655 ± 183 a	33 ± 3 b	19 ± 8 a		52 ± 1 b	-52 ± 1 c
C1P1	93 ± 3 a	38 ± 4 a	1706 ± 88 a	1663 ± 97 a	40 ± 5 a	16 ± 5 ab	150	56 ± 2 ab	94 ± 2 a

440 Abbreviations: C0/C1: Silty loam soil without/with long-term compost application, P0/P1: Silty loam soil without/with P fertilization.

441 ⁽¹⁾ For M1 +M2

442 ⁽²⁾ Each pot contains 4 kg soil, data per pot divided by 4

443 ⁽³⁾ For the entire plant (shoot + root),

444 ⁽⁴⁾ Fertilizer kg⁻¹ added

445 ⁽⁵⁾ calculated as the total P removed (plant P uptake of shoots and roots)

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448 **Table 3** Overall means and significances determined by a 3 WAY ANOVA analysis over the analysed variables (Plant dry matter (g pot⁻¹), plant P
449 concentration (mg kg⁻¹), plant P uptake (mg kg⁻¹) and soil P fractions (mg kg⁻¹). Different letters indicate significant differences between the
450 treatments within the main factors ($\alpha = 0.05$, Duncan's multiple range test).

	plant dry matter (g pot ⁻¹) ⁽¹⁾		plant P concentration (mg kg ⁻¹)		plant P uptake (mg kg ⁻¹) ⁽²⁾		soil P fractions (mg kg ⁻¹)						
	Shoot	Root	Shoot	Root	Shoot	Root	NaHCO ₃ -P _i	NaHCO ₃ -P _o	NaOH-P _i	NaOH-P _o	H ₂ SO ₄ -P _i	H ₂ SO ₄ -P _o	CAL-P
Overall means													
C0	85 b	41 a	1520 a	1518 b	32 b	16 a	136 b	30 b	162 a	31 a	248 b	48 a	130 b
C1	93 a	42 a	1562 a	1659 a	35 a	18 a	178 a	42 a	158 a	26 a	311 a	64 a	217 a
P0	86 a	44 a	1352 b	1459 b	29 b	16 a	114 b	28 b	145 b	37 a	271 a	55 a	137 b
P1	91 a	40 a	1731 a	1718 a	34 a	17 a	200 a	44 a	175 a	21 b	289 a	56 a	210 a
M0	--	--	--	--	--	--	183 a	36 b	161 b	62 a	278 a	36 c	257 a
M1	35 b	23 a	1632 a	1765 a	58 b	40 a	155 b	27 c	197 a	21 b	288 a	51 b	115 c
M2	53 a	19 b	1451 b	1413 b	78 a	27 b	133 c	45 a	122 c	4 c	274 a	80 a	148 b
P-values													
C x P	0.246	0.174	0.023	0.000	0.030	0.001	0.918	0.000	0.918	0.165	0.454	0.054	0.124
C x M	0.120	0.099	0.014	0.728	0.004	0.023	0.054	0.217	0.624	0.000	0.137	0.307	0.100
P x M	0.615	0.359	0.747	0.078	0.037	0.023	0.000	0.035	0.166	0.000	0.155	0.607	0.748
C x P x M	0.558	0.132	0.040	0.415	0.107	0.023	0.178	0.805	0.004	0.000	0.481	0.635	0.955

Abbreviations: C0/C1: Silty loam soil without/with long-term compost application, P0/P1: Silty loam soil without/with P fertilization, M0/M1/M2: before/after 1st cycle/after 2nd cycle.

⁽¹⁾ For M1 +M2

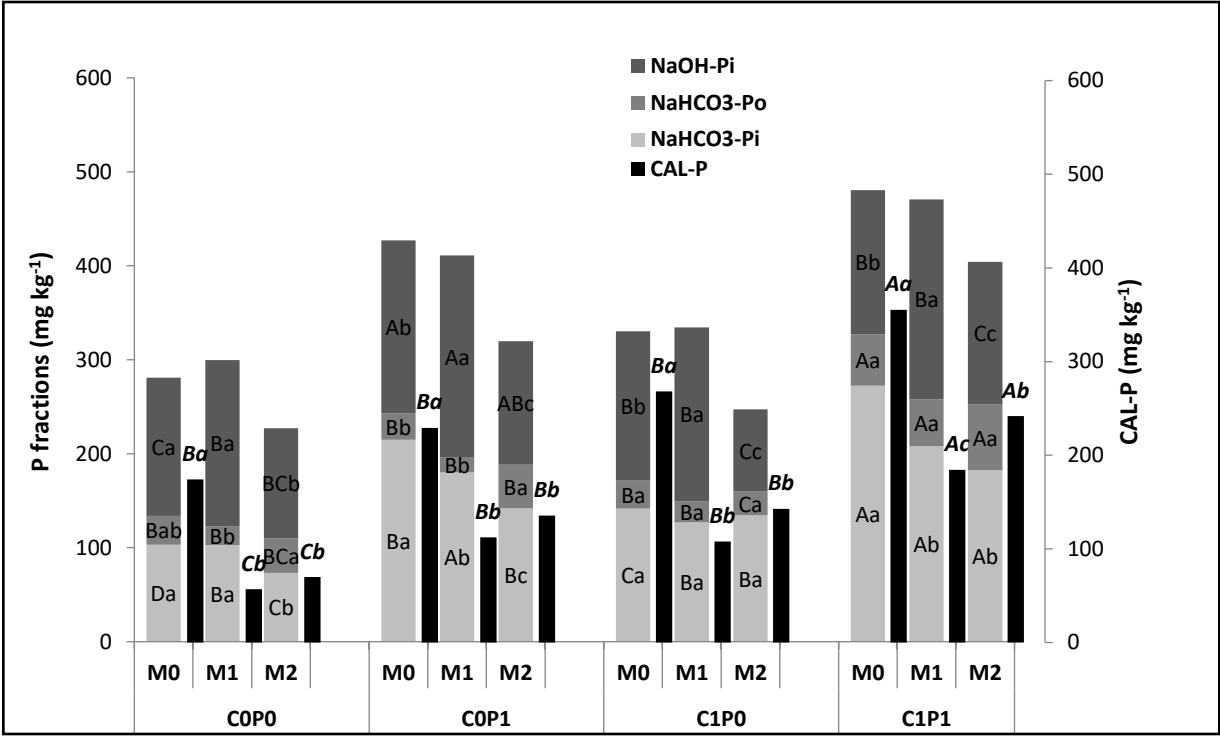
⁽²⁾ Each pot contains 4 kg soil, data per pot divided by 4.

Table 4 Pearson correlations between the plant P uptake (shoots and roots) and P fractions as determined before and after its respective cropping cycle (data from M1 and M2) and between the P balance and P fractions.

	P-CAL	NaHCO ₃ -P _i	NaHCO ₃ -P _o	NaOH-P _i	NaOH-P _o	H ₂ SO ₄ -P _i	H ₂ SO ₄ -P _o	Labile P ⁽¹⁾
Before cycle P fractions								
Plant P uptake								
shoots	-0,14	0,42*	0,06	0,74**	-0,12	0,28	0,01	0,60**
roots	0,58**	0,28	0,17	-0,38*	0,04	0,11	-0,23	0,12
sum	0,23	0,57**	0,16	0,44*	-0,03	0,34	-0,13	0,63**
After cycle P fractions								
shoots	0,66**	0,37	0,59**	-0,23	-0,88**	0,16	0,36	0,20
roots	-0,05	0,27	-0,33	0,50**	-0,51**	0,16	-0,23	0,35
sum	0,59**	0,51**	0,36	0,11	-0,87**	0,22	0,18	0,41*
P balance								
input	0,38	-0,86**	0,71**	0,35	-0,69**	0,43	0,25	-0,29
output	0,11	-0,52*	0,26	-0,02	-0,24	0,50	0,31	-0,42
balance	0,40	-0,87**	0,74**	0,38	-0,71**	0,41	0,23	-0,26

*,** significant at P < 0.05 and P < 0.01, respectively.

⁽¹⁾ Labile P calculated as NaHCO₃-P_i + NaHCO₃-P_o + NaOH-P_i



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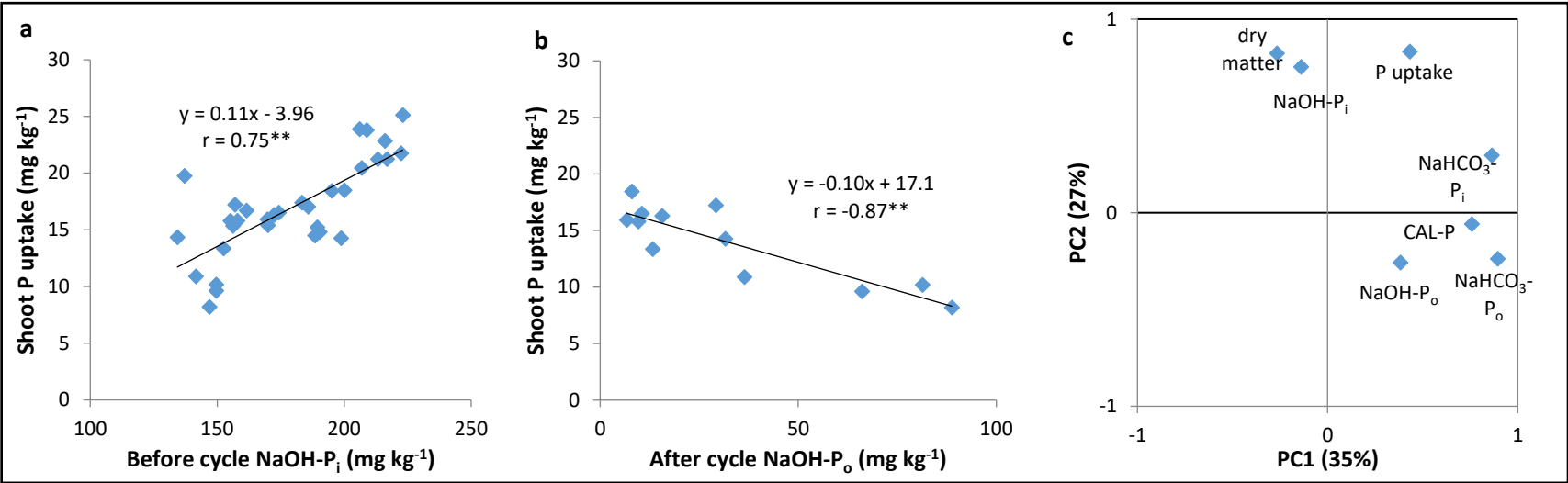
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Fig. 1 Distribution of P between soil fractions and CAL-P as determined for the experimental treatments (compost application x fertilizer application) at 4 days after soil fertilization (M0), and after 1st (M1) and 2nd (M2) maize harvest. C0/C1 indicate without/with long-term compost application, P0/P1 indicate without/with P fertilization. Different upper-case letters and lower-case letters in the same P fraction indicate statistical differences between different treatments at the same sampling time and between different sampling times within each treatment, respectively, determined by Duncan's Multiple Range Test (P< 0.05).

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Fig. 2 Relationships between a) P uptake and NaOH- P_i concentration in soil as determined before its respective maize cycle, b) P uptake and NaOH- P_o concentration in soil as determined after cycle (both with data from M1 and M2), and c) Principal component analysis with initial soil P fertility parameters and dry matter and P uptake data from M1+M2. *,** significant at $P < 0.05$ and $P < 0.01$, respectively.

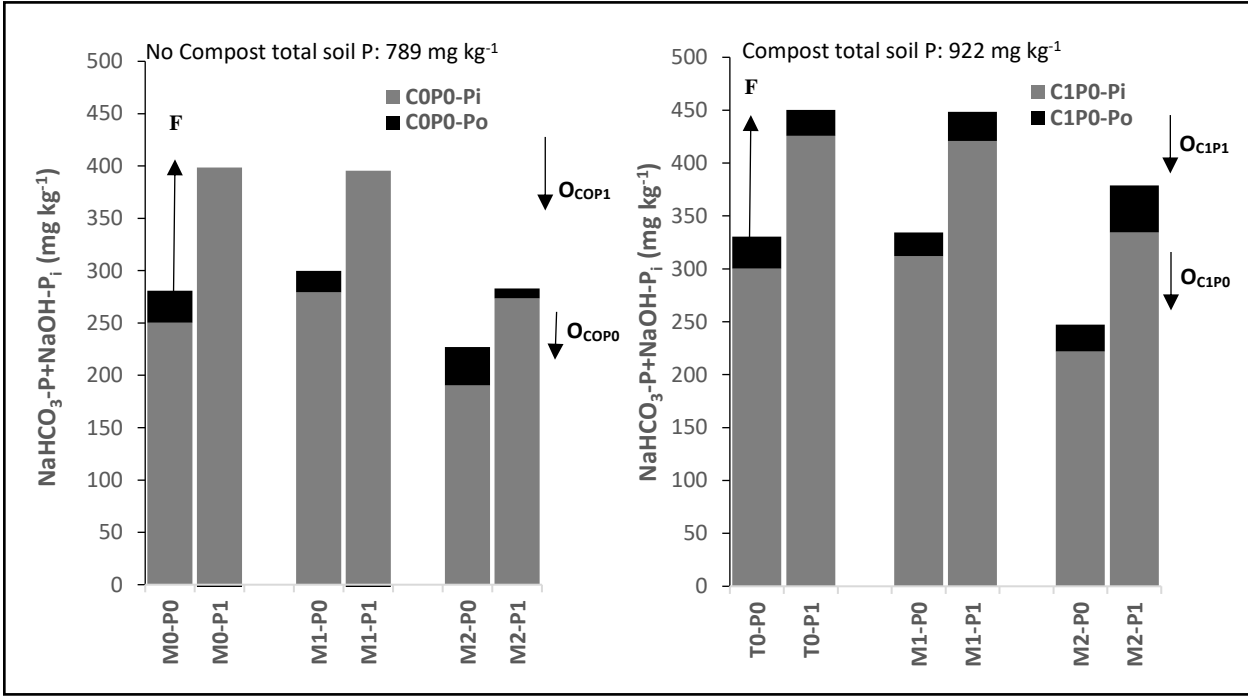


Fig. 3 Increment of labile + relative labile P_i ($\text{NaHCO}_3\text{-P}_i + \text{NaOH-P}_i$) and P_o ($\text{NaHCO}_3\text{-P}_o$) between the not fertilized soil (COP0 and C1P0) and the fertilized soils (with C1P0 and C1P1 showing the increment of P). M0, M1 and M2 represent the different sampling times and the arrows represents the amounts of P imported to the soil due fertilization and incorporated by plant biomass (equivalent to plant P uptake) in no compost (2a) and compost added soil (2b). **F**: Fertilization input= 150 mg P kg^{-1} in COP1 as in C1P1; O_{COP0} , O_{COP1} , O_{C1P0} , O_{C1P1} : P outputs of treatments COP0, COP1, C1P0 and C1P1, respectively. Length of the arrows represents magnitude of P outputs.