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Labile carbon facilitated phosphorus solubilization as regulated by bacterial and fungal communities in *Zea mays*

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ABSTRACT

Organic carbon (C) is often applied to agricultural soils to increase soil organic matter, however, its mechanistic effects on soil P transformations and availability resulting from stimulation of microbial activities and changes in microbial communities remain uncertain. This study investigated the responses of soil P availability, P fractions and *phoD* harboring bacterial and fungal communities to two dose rates (5 and 10 mg C g^{-1} dry soil) of labile C (glucose) in bulk and rhizosphere soils planted with maize in a P-deficient soil (Oxisol). Both doses of glucose significantly increased available P concentrations (over a 47-day period) in soils without maize, and in the bulk and rhizosphere soils of maize, resulting in the promotion of maize growth. Glucose additions altered soil *phoD* harboring bacterial and fungal community composition and stimulated the growth of keystone P-solubilizing microorganisms, such as *Bradyrhizobium* and *Eupenicillium*, in soils with maize. Co-occurrence network analysis showed that glucose enhanced interactions between *phoD* harboring bacterial taxa relative to that of fungal taxa, whether in individual networks or in combined networks. Our results highlight the importance of labile C in facilitating changes in soil P-solubilizing bacterial and fungal communities of a P-deficient soil. These findings provide crucial information to guide P-cycling management strategies via microbial regulation in agro-ecosystems.

1. Introduction

Phosphorus (P) plays an important role in plant nutrition as a component of many important biochemical compounds (*e.g.,* ATP, phospholipid, DNA, RNA) (George et al., 2018). Phosphate fertilizer is commonly applied to alleviate P deficiency in croplands, but it may become strongly bound into non-labile soil fractions through a series of biogeochemical reactions. Long-term application of P fertilizer has resulted in high concentrations of total P in soils, but often low P availability as much of the bound mineral P is not accessible to plants and microorganisms (Kochian, 2012; Václavková et al., 2018). Thus, plant growth is often limited by soil P deficiency, especially in strongly P-fixing soils (*e.g.*, Oxisols, Andisols). Therefore, developing effective strategies to mobilize the accumulated P in soils is strongly warranted to sustain crop yields and soil health.

Soil microorganisms control key biogeochemical processes in agro-ecosystems and play a crucial role in nutrient cycling and energy flows (Fierer, 2017; Dai et al., 2018, 2020; Ma et al., 2021). Bacteria and fungi strongly regulate soil P availability through solubilization (release of soluble P from mineral P), mineralization (decomposition of organic P) and immobilization (incorporation of P in microbial biomass) processes (Richardson and Simpson, 2011). Phosphorus solubilizing bacteria promote soil P availability by direct oxidation pathways, producing organic anions and protons, secreting siderophores and releasing phosphatase and cellulolytic enzymes (Sharma et al., 2013). Under P deficiency, certain microbes are observed to up-regulate the expression of P starvation-induced genes and produce various phosphatases (*e.g.,* alkaline phosphatase, phytase, C–P lyase) (Nannipieri et al., 2011) to

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promote mineralization of organic P. Alkaline phosphatase is encoded by three homologous genes (*phoA*, *phoD*, *phoX*) produced by microorganisms (Kathuria and Martiny, 2011). Due to its prominence throughout bacterial communities and across soil types, the *phoD* gene is considered to be the most critical alkaline phosphatase-encoding gene (Tan et al., 2013; Ragot et al., 2015). The abundance and community composition of *phoD* harboring bacteria are sensitive to soil P status and play a vital role in mediating P transformations.

Fungi are eukaryotic microorganisms that commonly function as decomposers, mutualists and pathogens to mediate nutrient cycling in terrestrial ecosystems (Tedersoo et al., 2014). Fungi can effectively decompose recalcitrant organic matter along with more labile organic matter pools, thereby playing an important role in soil nutrient mobilization and immobilization (Krauss et al., 2011; de Vries and Caruso, 2016). The typical filamentous structure of most fungi facilitates their penetration into organic matter particles to obtain nutrients from soil materials (Fabian et al., 2017). At present, about 80% of terrestrial plant species are believed to form symbiotic associations with mycorrhizal fungi, with enhance P uptake being a prominent synergy (Smith and Smith, 2011). Thus, pursuing management strategies to increase soil P availability through promoting bacterial and fungal activities/interactions has compelling applications in sustainable agriculture.

Inputs of organic substrates can drive changes in soil microbial communities, with corresponding changes in soil functions. For example, carbohydrate (*e.g.,* glucose, glucose-6-phosphate, cellulose) application increased C assimilation within microbial biomass and total PLFA between 0 and 20 days, while slurry application stimulated respiration and increased fungal PLFA in an agricultural grassland soil (Abbruzzese et al., 2021). Moreover, a 16S rRNA gene-based pyrosequencing and microarray-based GeoChip revealed that long-term application of rice straw, with or without chemical fertilizers, shifted the bacterial community composition and increased the diversity and abundance of C/N cycling genes in a paddy soil (Ding et al., 2018). Microorganisms are posited to exhibit a relatively stable C:N:P stoichiometry (Cleveland and Liptzin, 2007) and can survive in soils with very low P availability, partly due to their ability to release P from insoluble sources (Kishore et al., 2015; He et al., 2021). Thus, the response of bacterial and fungal communities to organic C-substrate additions may increase soil P availability.

Root exudates provide abundant nutrients and energy for rhizosphere microorganisms, which promote higher chemical and biological activities in the rhizosphere versus bulk soil (Frey, 2007). The biological interactions and biogeochemical processes that occur in the rhizosphere affect the dynamics of P uptake by plants (Richardson, 2001). Microbes can enhance the availability of nutrients (such as P) for plant uptake via altering rates of nutrient supply and resource partitioning, further stimulating plant productivity (van der Heijden et al., 2008). For example, arbuscular mycorrhizal fungi contributed up to 90% of plant P uptake in a pot study using single or multiple plant species (Munkvold et al., 2004; van der Heijden et al., 2006). Inputs of organic C may facilitate the growth of microbes, thereby affecting plant growth and P uptake. Long-term labile C additions (*e.g.,* manure) increased concentrations of labile and moderately-labile P in a paddy soil by increasing carbon inputs and altering soil C:N:P stoichiometry, which indirectly promoted the uptake of P by rice (Qaswar et al., 2019). Moreover, green manure and rice straw increased the phosphorus solubilizing microbial population, soil enzyme activity and microbial biomass, thereby promoting P transformations and increasing rice yield (Chatterjee et al., 2021).

To gain a mechanistic understanding of the effects of labile C sources on P availability to plants, we examined the effects of two dose rates of glucose on soil P availability, P transformations and the *phoD* harboring bacterial and fungal community composition in the bulk and rhizosphere soils of maize growing in a P-deficient Oxisol. We hypothesized that i) glucose addition will increase P availability and its release rate, and alter the composition of P fractions in both bulk and rhizosphere soils; ii) glucose additions will change the *phoD* harboring bacterial and fungal community structure, thereby enhancing the growth of specific taxa that facilitate P release in the maize rhizosphere; and iii) the soil *phoD* harboring bacterial community will contribute more to P solubilization than the fungal community in the P-deficient soil. Our findings provide a mechanistic insight into the relationship between crop residue management and P supply with direct application to sustainable agricultural practices.

2. Materials and methods

2.1. Site description

The experimental soil was classified as an Oxisol in Soil Taxonomy (United States Department of Agriculture). We collected the topsoil (0–20 cm) from an upland agricultural field at Jiangxi Institute of Red Soil in Jiangxi Province, China (116°20′24″E, 28°15′30″N). The soil was air-dried, homogenized and sieved (<2 mm) for subsequent pot experiments. Basic soil properties were: pH 5.30, total C 7.9 g kg^{-1}, total N 0.8 g kg^{-1}, total P 584 mg kg^{-1}, available P 6.7 mg kg^{-1} (Bray-P), water holding capacity (water content at −0.03 MPa) 18.9%, and clay loam texture (clay 34.6%, silt 39.4%, sand 26.1%).

2.2. Pot experiment

Deionized water was added to the prepared soil to reach 80% water holding capacity, and pre-incubated at 25 °C for one week. The pot experiment included three treatments with three replicates: (1) CK, no glucose addition; (2) G5, 5 mg glucose-C g^{-1} dry soil addition; and (3) G10, 10 mg glucose-C g^{-1} dry soil addition. All treatments were run with a secondary treatment of with or without maize (*Zea mays*; variety Zhengdan 958) cultivation. Thus, a total of eighteen (3 glucose levels × with/without maize × 3 replicates = 18) nursery pots (14.2 cm diameter, 17.3 cm height) were filled with homogenized soil (equivalent to 2.6 kg dry soil). For treatments with maize cultivation, nylon bags (48-μm mesh, 10 cm × 15 cm) were used to exclude roots and separate bulk soil from the rhizosphere. The soil mass was equally distributed inside and outside of the bag. The mesh size of the nylon bags was small enough to allow only soluble nutrients to pass through. Rhizosphere soils were collected as soil attached to the roots; bulk soils were collected as far away from the mesh bag as possible to avoid the effects of root activity. A total of 120 mg K kg^{-1} (KCl) and 200 mg N kg^{-1} (NH_4NO_3) were applied to each pot as basal fertilizers. Three germinated maize seeds were sown in the center of the pot, grown in a greenhouse at 28 °C with a 12/12 h light/dark cycle and maintained a constant water content with the weighing method. After growing three days, a single seedling with the same growth/development was retained in each pot. We collected soils without maize, and bulk/rhizosphere soils and plant aboveground (stems/leaves)/belowground (roots) components from maize treatments for a series of analyses after 47 days of growth (tenth-to fourteen-leaf stage).

2.3. Soil biochemical properties and plant physicochemical analyses

Soil pH (soil-to-water ratio of 1:2.5, v/v) was measured with a pH meter (PHSJ-3F, INESA Scientific Instrument, China). Available P (Bray-P) was extracted with 0.03 M NH_4F/0.025 M HCl and quantified using the molybdenum blue method (Murphy and Riley, 1962). Microbial biomass C (MBC) and P (MBP) were measured by the $CHCl_3$ fumigation-extraction method (Brookes et al., 1982). Soil potential acid phosphatase (ACP) and alkaline phosphatase (ALP) activities were determined using the p-nitrophenyl phosphate method (Tabatabai, 1994). The reaction mixture consisted of 1 g fresh soil, 0.2 mL toluene, 4 mL buffer (composed of 0.1 M Tris(hydroxymethyl)methyl aminomethane, 0.1 M maleic acid, 0.07 M citric acid, 0.1 M boric acid and 0.5 M NaOH, adjust pH to 6.5 and 11.0 with 1 M HCl/NaOH for ACP/ALP

activities, respectively) and 1 mL p-nitrophenyl phosphate disodium solution. Reacting suspensions were mixed thoroughly and incubated at 37 °C for 1 h. After incubation, 1 mL 0.5 M $CaCl_2$ and 4 mL 0.5 M NaOH were added and enzyme activity was measured using a colorimetric method at 410 nm (Tabatabai, 1994).

Fresh plant materials were oven-dried at 105 °C for 30 min, then at 65 °C to obtain a constant weight. Plant aboveground and belowground biomass was measured gravimetrically (±1 mg) immediately after drying. Plant tissues were ball-milled to a homogenous fine powder for total P analysis by digestion with H_2SO_4–H_2O_2 and quantification using the molybdenum blue method. Plant absorbed P was calculated from the plant biomass and total P concentration.

2.4. Soil P fractions

Soil P fractions were determined using a sequential extraction procedure described by Tiessen and Moir (1993), as modified from Hedley et al. (1982). In brief, 0.5 g sieved dry soil (<0.15 mm) was sequentially extracted with 30 mL MilliQ water, 0.5 M $NaHCO_3$ (pH 8.5), 0.1 M NaOH and 1 M HCl in 50 mL centrifuge tubes (polypropylene) on a reciprocating shaker (200 rpm) at 25 °C for 16 h. The extracts were collected by centrifuging the soil suspension at 10,000 g for 10 min at 0 °C. The molybdenum blue method was used to quantify each extract for inorganic P (P_i) and total P following digestion with H_2SO_4–$HClO_4$. Organic P (P_o) was calculated as the difference between total P and inorganic P. Organic P in the H_2O extract was not detectable (<1 mg P kg^{-1}). The residual P fraction was calculated as the difference between total P and the sum of sequentially extracted P fractions. The H_2O-P_i and $NaHCO_3$-P_i fractions represent soluble and weakly adsorbed inorganic P; $NaHCO_3$-P_o represents easily mineralizable organic P; NaOH-P_i and NaOH-P_o represent intermediately labile inorganic/organic P; and HCl-P_i, HCl-P_o and residual-P represent non-labile P forms.

2.5. DNA extraction and qPCR analysis

Total DNA was extracted from 0.5 g fresh soil using the Fast DNA® SPIN Kit for Soil (QBIOGENE, Carlsbad, CA, USA) following manufacturer's instructions. Concentrations and quality of DNA samples were determined using a NanoDrop® ND-2000 spectrophotometer (NanoDrop Technologies, Wilmington, DE, USA). The extracted DNA was stored at −20 °C for subsequent assays.

The prepared DNA was amplified with F730 (5′-CAGTGGGACGACCACGAGGT-3′) and R1101 (5′-GAGGCCGATCGGCATGTCG-3′) (Sakurai et al., 2008) using real-time quantitative PCR on a LightCycler®480II (Roche, Germany) to quantify *phoD* gene abundance. Reactions were performed in a 25 μL mixture containing 12.5 μL SYBR®Premix Ex Taq II (Takara, Japan), 0.25 μL PCR forward and reverse primers (both 50 μM), 2 μL DNA template and 10 μL double distilled water (ddH_2O). Amplification conditions were as follows: 1 cycle at 95 °C for 30 s, 40 cycles at 95 °C for 5 s, 59 °C for 30 s and 72 °C for 30 s. The plasmid containing target gene was serially diluted 10 times to span 10^3 to 10^9 copies μL^{-1} for used as the standard curve. Amplification efficiencies for the *phoD* gene were 85–92% with R^2 values > 0.99.

2.6. Determination of phoD harboring bacterial and fungal community composition using Illumina sequencing

The *phoD* gene amplification was conducted with F730/R1101 primers. Thermal conditions were as follows: 98 °C for 5 min; 35 cycles at 98 °C for 30 s, 59 °C for 30 s and 72 °C for 45 s; and 72 °C for 5 min. The fungal ITS1 region was amplified with primers ITS5F (5′-GGAAGTAAAAGTCGTAACAAGG-3′) and ITS2R (5′-GCTGCGTTCTTCATCGATGC-3′) (White et al., 1990). Thermal conditions were as follows: 98 °C for 5 min; 28 cycles at 98 °C for 30 s, 52 °C for 45 s and 72 °C for 45 s; and 72 °C for 5 min. PCR products were sequenced on the Illumina HiSeq PE250 sequencing platform (Illumina, San Diego, CA, USA). The raw sequence data reported in this paper were deposited in the Genome Sequence Archive (GSA) database (accession number CRA003873).

Raw reads were merged by USEARCH v11.0 (Edgar, 2010), quality-filtered and singletons removed by VSEARCH 2.14.1 (Rognes et al., 2016). The remaining sequences were clustered into operational taxonomic units (OTUs) with ≥75% similarity (Tan et al., 2013; Fraser et al., 2015; Wei et al., 2019) for *phoD* harboring bacteria and ≥97% similarity for fungi. Chimeras were filtered using the UPARSE algorithm (Edgar, 2013), then *phoD* harboring bacterial OTU taxonomic classification was conducted using BLAST; fungi were annotated in the RDP v16.0 database. Bacterial sequencing data were rarefied to a minimum sequencing depth of 78,000 reads, and fungal sequencing data to a depth of 60,000 reads.

2.7. Statistical analyses

One-way ANOVA followed by a least significant difference (LSD) test was performed to examine differences of soil biogeochemical properties, plant biomass and absorbed P among treated soils using SPSS 20.0. Bray-Curtis based principal components analysis (PCoA) and redundancy analysis (RDA) were performed using vegan package in R 3.6.3. Similarities and differences in glucose addition treatments were evaluated by analysis of similarities (ANOSIM) using vegan package in R 3.6.3. Microbial co-occurrence networks of *phoD* harboring bacteria, fungi and combined *phoD* harboring bacteria and fungi for treated soils with maize were built using igraph package in R 3.6.3 and visualized using Gephi 0.9.2. The differences between glucose-treated genera and control were assessed and plotted with Statistical Analysis of Metagenomic Profiles (STAMP 2.1.3) (Parks et al., 2014). Correlation analysis among soil properties was determined using corrplot package in R 3.6.3.

3. Results

3.1. Plant biomass, total P concentration of biomass and plant absorbed P

After 47 days of maize growth, plant biomass and total P concentrations in aboveground and belowground biomass differed among the three treatments (Fig. S1). The G5 treatment significantly increased maize aboveground biomass by 0.10 g pot^{-1}; G10 treatment increased 0.19 g $pot^{-1}$and 0.07 g pot^{-1} for aboveground and belowground components, respectively (Fig. S1a). Glucose additions did not affect plant total P concentration in aboveground biomass; however, the G10 treatment increased plant total P concentration in belowground biomass (Fig. S1b). Correspondingly, the amount of P absorbed by the maize in G5 treatment increased 0.07 mg $plant^{-1}$ for aboveground biomass, and the G10 treatment increased 0.13 mg $plant^{-1}$ and 0.08 mg $plant^{-1}$ for aboveground and belowground biomass, respectively ($p < 0.05$) (Fig. 1a).

3.2. Soil pH, available P and P fractions

Significant differences ($p < 0.05$) in glucose treated soil pH were observed among the three treatments (Fig. S1c). Compared with CK, glucose additions significantly increased pH by 0.15–0.7 units in all glucose treated soils (except for the G5-rhizosphere soil). Glucose additions significantly affected soil P availability, release rate of available P and various P fractions in both soils without maize and with maize (bulk soil and rhizosphere) (Fig. 1b and c and Table 1). Both G5 and G10 treatments increased available P concentrations in soils without maize (increase of 1.3 mg kg^{-1} with a 0.025 mg kg^{-1} d^{-1} release rate for available P in G5 treatment and 3.1 mg kg^{-1} with a 0.064 mg kg^{-1} d^{-1} release rate for available P in G10 treatment) and bulk soil (5.2 mg kg^{-1} with a 0.11 mg kg^{-1} d^{-1} release rate for available P in G5 treatment and 7.2 mg kg^{-1} with a 0.15 mg kg^{-1} d^{-1} release rate for available P in G10 treatment) ($p < 0.05$) (Fig. 1b). With respect to the rhizosphere soil of maize, the G10 treatment increased P availability by 1.7 mg kg^{-1}, but

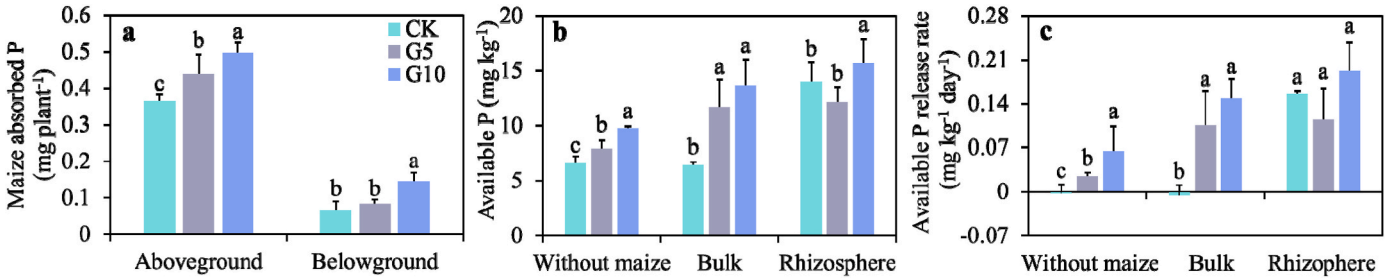

Fig. 1. Maize absorbed P (a), concentrations of available P (b) and its release rate (c) among three soil treatments (CK, no glucose addition; G5, 5 mg glucose-C g^{-1} dry soil addition; G10, 10 mg glucose-C g^{-1} dry soil addition) without maize and with maize (bulk soil and rhizosphere). Available P release rate calculated as the change in available P concentration divided by the incubation time. Error bars indicate the standard deviation of triplicate analyses. Different letters indicate significant differences among treatments ($p < 0.05$).

Table 1
Hedley sequentially-extracted P fractions (mg kg^{-1}) among three soil treatments (CK, no glucose addition; G5, 5 mg glucose-C g^{-1} dry soil addition; G10, 10 mg glucose-C g^{-1} dry soil addition) without maize and with maize (bulk soil and rhizosphere) (mean ± standard deviation, n = 3). Different letters indicate significant differences among treatments ($p < 0.05$). H_2O-P_i, H_2O extracted inorganic P; $NaHCO_3$-P_i/P_o, $NaHCO_3$ extracted inorganic/organic P; NaOH-P_i/P_o, NaOH extracted inorganic/organic P; HCl-P_i/P_o, HCl extracted inorganic/organic P; and Residual-P, remaining P after all extractions.

Soil	Treatment	H_2O-P_i	$NaHCO_3$-P_i	$NaHCO_3$-P_o	NaOH-P_i	NaOH-P_o	HCl-P_i	HCl-P_o	Residual-P
Without maize	CK	4.0±1c	16±2b	41 ± 10a	199 ± 11a	90±8b	17±1a	3±3c	212 ± 12a
	G5	13±1b	25±4 ab	33±2a	179 ± 10b	103±2 ab	17±2a	10±3b	203 ± 16a
	G10	10±1a	27±7a	32±9a	171±6b	110 ± 14a	18±1a	19±2a	196 ± 37a
Bulk soil with maize	CK	2.0 ± 0.6b	30±3a	20±2b	197 ± 12a	95±6b	17±3a	29 ± 10a	194 ± 33a
	G5	6.1 ± 1.3a	24±2a	28±1a	178±5b	112 ± 11a	19±3a	12±1b	205 ± 21a
	G10	4.9 ± 1.1a	28±3a	31±6a	178±9b	115±8a	21±3a	17±1b	189 ± 10a
Rhizosphere with maize	CK	0.9 ± 0.2b	29±3a	24±8b	198±8a	99±8a	20±2a	18±2a	195 ± 16a
	G5	1.7 ± 0.5b	24±3a	31±3 ab	179±7b	107±4a	17±3a	15±4a	209 ± 16a
	G10	5.4 ± 1.7a	26±2a	38±2a	175±8b	113±9a	18±2a	17±1a	192 ± 19a

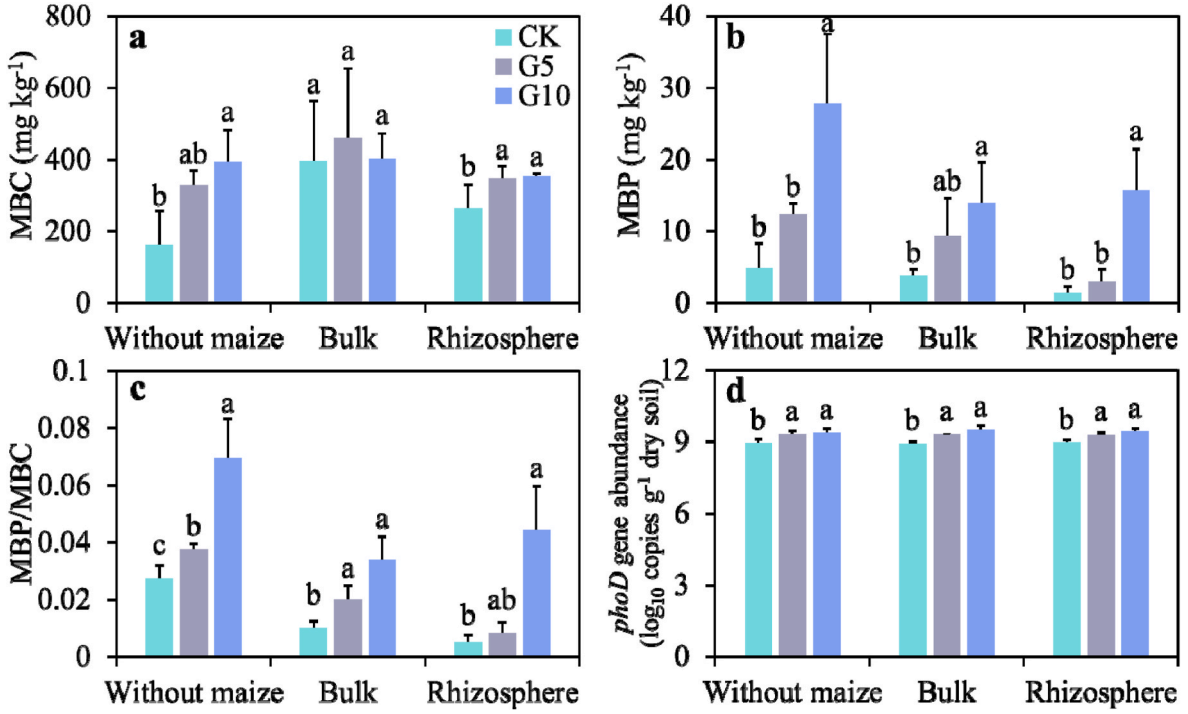

Fig. 2. Concentrations of microbial biomass carbon (MBC) (a) and phosphorus (MBP) (b), MBP/MBC ratio (c) and *phoD* gene abundance (d) among three soil treatments (CK, no glucose addition; G5, 5 mg glucose-C g^{-1} dry soil addition; G10, 10 mg glucose-C g^{-1} dry soil addition) without maize and with maize (bulk soil and rhizosphere). Error bars indicate the standard deviation of triplicate analyses. Different letters indicate significant differences among treatments ($p < 0.05$).

there was no effect for the G5 treatment (Fig. 1b).

Among various P fractions in soils without maize, both G5 and G10 treatments increased the concentrations of H_2O-P_i and HCl-P_o, whereas the concentrations of NaOH-P_i decreased. In soils with maize, the bulk soils of G5 and G10 treatments showed increased concentrations of H_2O-P_i, $NaHCO_3$-P_o and NaOH-P_o, and decreased concentrations of NaOH-P_i and HCl-P_o. Rhizosphere soils of maize responded to the G5 and G10 treatments with decreased concentrations of NaOH-P_i, whereas the G10 treatment also had increased concentrations of H_2O-P_i (Table 1).

3.3. Microbial biomass and potential ACP/ALP activities

Microbial biomass C (MBC), microbial biomass P (MBP) and the MBP/MBC ratio responses to glucose additions are shown in Fig. 2. After 47 days of maize growth, the G5 treatment did not affect MBC and MBP concentrations ($p > 0.05$), but increased the MBP/MBC ratio of soil without maize and the bulk soil of maize ($p < 0.05$). G10 treatments significantly increased MBC and MBP concentrations and the MBP/MBC ratio in all treated soils, except for MBC in bulk soil ($p < 0.05$). In all glucose treated soils, the G5 and G10 treatments significantly enhanced potential ACP and ALP activities (Table 2).

3.4. Soil phoD gene abundance and phoD harboring bacterial and fungal community composition

In comparison to the CK treatment, higher *phoD* gene abundances were observed in all G5 and G10 treatments (Fig. 2d). G5 and G10 treatments increased *phoD* gene abundance by 1.2×10^9 and 1.6×10^9 copies g^{-1} dry soil in soils without maize, respectively. Similarly in soils with maize, the G5 and G10 treatments increased *phoD* gene abundance by 1.2×10^9 and 2.2×10^9 copies g^{-1} dry soil in bulk soil, and by 1.1×10^9 and 1.8×10^9 copies g^{-1} dry soil in rhizosphere soil, respectively ($p < 0.05$). ANOSIM showed that both G5 and G10 treatments changed the composition of *phoD* harboring bacterial and fungal communities compared to CK treatment ($p < 0.05$) (Fig. S2 and Table S1). Statistical analysis of metagenomic profiles (STAMP) revealed the specific genera of *phoD* harboring bacterial and fungal communities responding to glucose additions ($p < 0.05$) (Figs. 3 and 4). For the *phoD* harboring bacterial community, the G5 treatment (relative to CK) significantly increased the relative abundance of *Ralstonia* and decreased the relative abundance of *Burkholderia* in soil without maize, increased the relative abundances of *Amycolatopsis* and *Intrasporangium* in bulk soil of maize, and increased the relative abundances of *Bradyrhizobium* and *Amycolatopsis* in rhizosphere soil of maize (Fig. 3a, c and e). The G10 treatment (relative to CK) significantly increased the relative abundances of *Bradyrhizobium* and decreased the relative abundances of *Streptomyces* and *Burkholderia* in soil without maize and the bulk soil of maize, and decreased the relative abundances of *Burkholderia* in rhizosphere soil of maize (Fig. 3b, d and f).

For the fungal community, *Eupenicillium* was the predominant genus in G5 (relative abundance 84–94%) and G10 (relative abundance 75–90%) treatments of soils with and without maize (Fig. 4). Both G5 and G10 treatments significantly decreased the relative abundances of *Cochliobolus*, *Aspergillus*, *Mortierella* and *Penicillium* in soils without maize and the bulk soil of maize, and decreased the relative abundances of *Davidiella* and *Chaetomium* in the rhizosphere soil of maize (Fig. 4). Additionally, *phoD* gene abundance showed significant positive correlations with the relative abundances of *Amycolatopsis*, *Bradyrhizobium*, *Mesorhizobium*, *Pseudolabrys*, *Xanthomonas* and *Eupenicillium*, and negative correlations with the relative abundances of 18 genera (Fig. S3).

Redundancy analysis (RDA) revealed that soil pH, *phoD* gene abundance and potential ACP activity were significant predictors for the composition of the *phoD* harboring bacterial community ($p < 0.05$) (Fig. S4a). The first and second axes of the RDA explained 35.4% and 13.5% of the total variation observed in the soil *phoD* harboring bacterial community, respectively. Notably, potential ACP activity was the main factor explaining the structure of the fungal community ($p < 0.05$) (Fig. S4b). The first and second axes of RDA explained 80.7% and 11.6% of the total variation observed in the fungal community, respectively.

3.5. Microbial co-occurrence networks

Microbial co-occurrence networks of OTUs were established based on Spearman correlations for the soil samples with maize in the CK, G5 and G10 treatments (Fig. 5). Glucose additions increased the node, edge and module numbers of *phoD* harboring bacterial networks, and decreased the node, edge and module numbers of fungal networks (Table S2a). The highest number of genera (*e.g.*, *Bradyrhizobium* and *Amycolatopsis*) with increased abundance in glucose treated soils was observed in the *phoD* harboring bacterial network of the G10 treatment (28), followed by the G5 treatment (19) and CK treatment (10). Notably, *Bradyrhizobium* was involved in the keystone module of the *phoD* harboring bacterial network of the G10 treatment, but not in the CK treatment.

For the combined *phoD* harboring bacterial and fungal networks, fungal nodes dominated in the CK treatment, whereas bacterial nodes dominated in the G5 and G10 treatments (Table S2b). Glucose additions increased the proportion of bacteria-bacteria edges, and decreased the proportion of bacteria-fungi and fungi-fungi edges (Table S2b). The genera (*e.g.*, *Bradyrhizobium*, *Amycolatopsis* and *Eupenicillium*) with increased abundances in glucose treated soils were all involved in the combined networks of the CK treatment; however, *Eupenicillium* was not found in the G5 and G10 treatments.

Table 2
Soil potential acid phosphatase (ACP) activity and alkaline phosphatase (ALP) activity among three soil treatments (CK, no glucose addition; G5, 5 mg glucose-C g^{-1} dry soil addition; G10, 10 mg glucose-C g^{-1} dry soil addition) without maize and with maize (bulk soil and rhizosphere) (mean ± standard deviation, n = 3). Different letters indicate significant differences among treatments ($p < 0.05$).

Soil	Treatment	ACP (μg pNPP g^{-1} dry soil h^{-1})	ALP (μg pNPP g^{-1} dry soil h^{-1})
Without maize	CK	196 ± 47c	37±5b
	G5	898 ± 26a	101±9a
	G10	705 ± 35b	102 ± 10a
Bulk soil with maize	CK	235 ± 15b	48±5b
	G5	848 ± 48a	95±3a
	G10	755 ± 117a	88±8a
Rhizosphere with maize	CK	284 ± 61b	59±3b
	G5	899 ± 51a	98±5a
	G10	856 ± 65a	122 ± 24a

4. Discussion

4.1. P availability response to labile C addition

Labile C addition significantly increased P availability in soils without maize and with maize (except in G5-rhizosphere) and promoted plant growth after 47 days of maize growth (Figs. 1b and S1a). Immobilization and mobilization of P in soil proceed as a dynamic quasi-equilibrium. We observed 584 mg kg^{-1} total P in our experimental soil; hence, there is strong potential for the release of available P from the soil. Due to microbial processes for P solubilization, when soil available P was consumed by plant growth, it can be continuously replenished from the soil by microbial processes. Thus, the soil available P pool was not measurably depleted by plant growth. Additionally, roots exudates, such as citrate, fructose and sucrose, could increase P availability through competing with soil P for sorption sites on Fe/Al (hydr) oxides (Gerke, 2015). The labile C-induced increase in available P concentrations is consistent with several previous studies (Spohn and

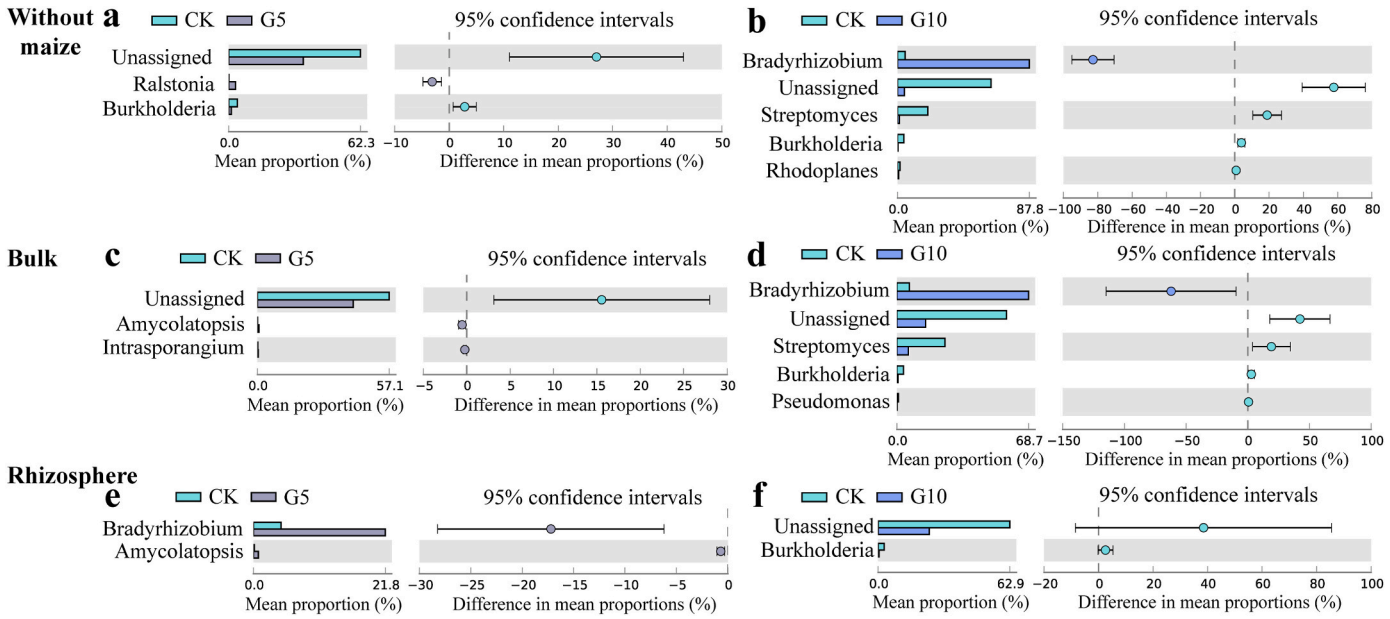

Fig. 3. Variation analysis of *phoD* harboring bacterial community in soils without maize (a and b), and with maize for bulk (c and d) and rhizosphere (e and f) soils with CK (no glucose addition) versus G5 (5 mg glucose-C g^{-1} dry soil addition) and CK (no glucose addition) versus G10 (10 mg glucose-C g^{-1} dry soil addition) at the genus level. Only genera with significant differences are shown ($p < 0.05$).

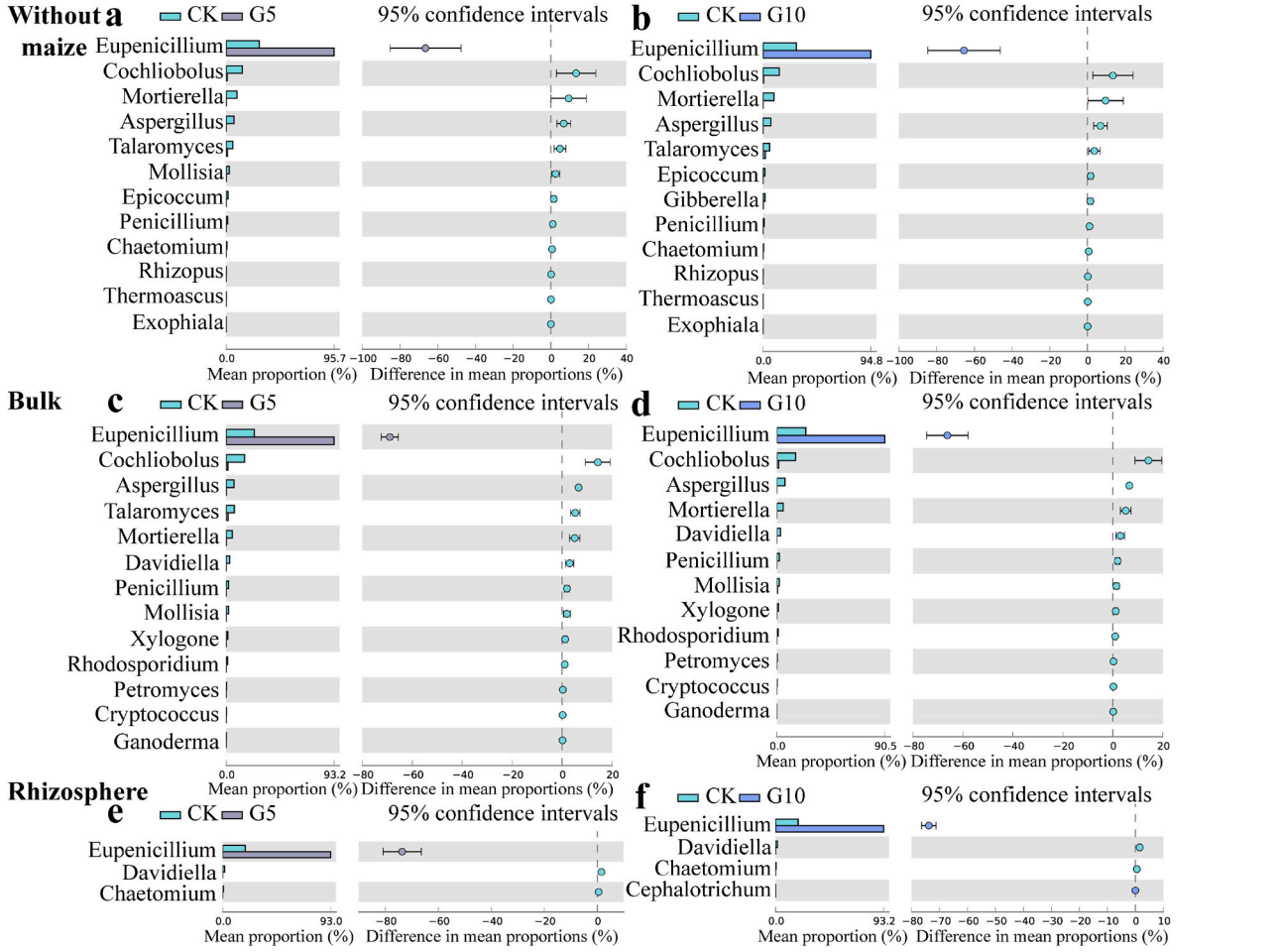

Fig. 4. Variation analysis of fungal community in soils without maize (a and b), and with maize for bulk (c and d) and rhizosphere (e and f) soils with CK (no glucose addition) versus G5 (5 mg glucose-C g^{-1} dry soil addition) and CK (no glucose addition) versus G10 (10 mg glucose-C g^{-1} dry soil addition) at the genus level. Only genera with significant differences are shown ($p < 0.05$).

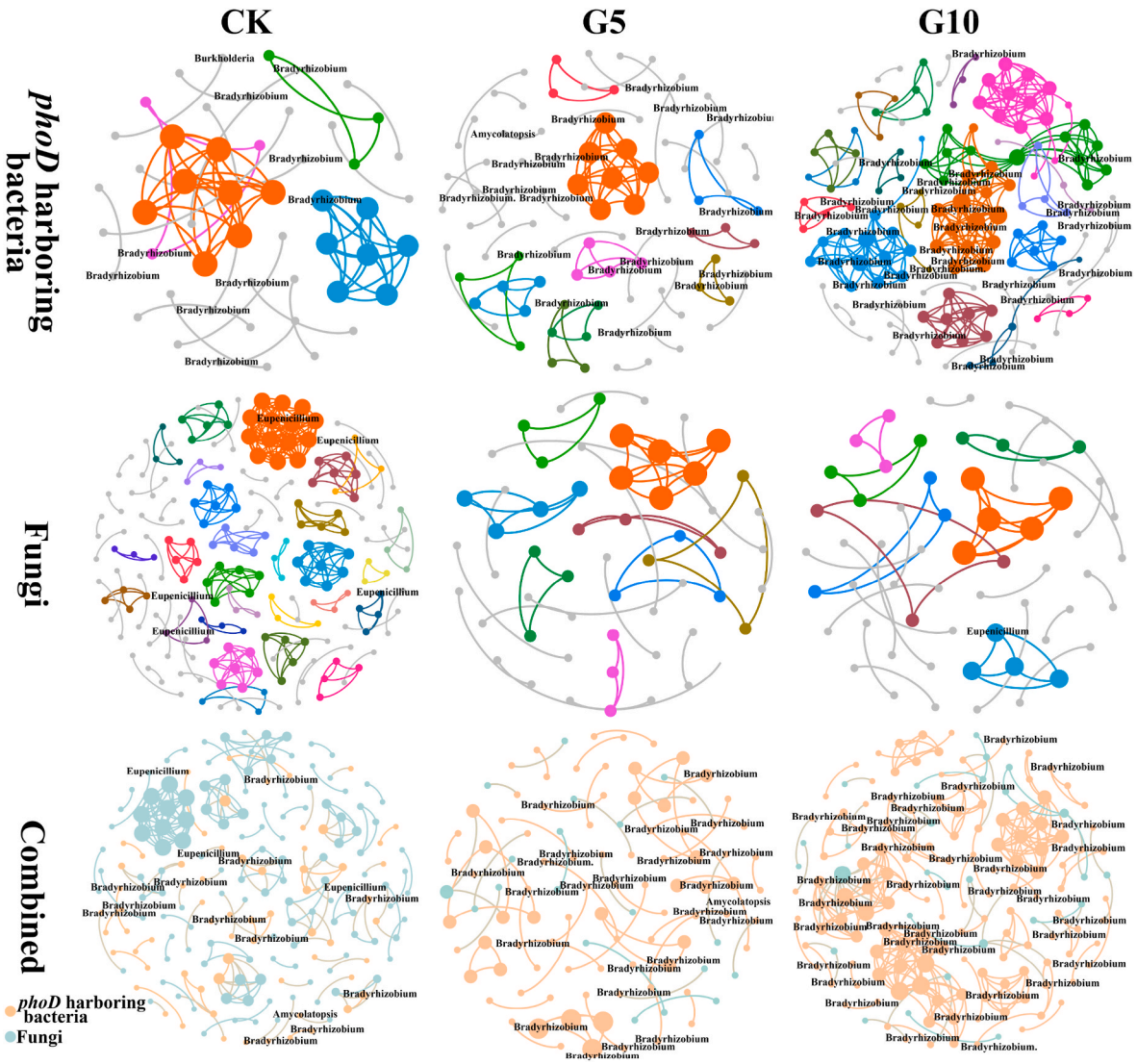

Fig. 5. Microbial co-occurrence networks of OTUs among three soil treatments (CK, no glucose addition; G5, 5 mg glucose-C g^{-1} dry soil addition; G10, 10 mg glucose-C g^{-1} dry soil addition) with maize. Node size indicates the degree of the node in all networks. Node color indicates modules in the *phoD* harboring bacterial and fungal networks, and indicate microbe types in the combined *phoD* harboring bacterial and fungal networks. Node labels indicate the increased genera of *phoD* harboring bacteria (*i.e.*, *Bradyrhizobium* and *Amycolatopsis*) and fungi (*i.e.*, *Eupenicillium*) in G5 and G10 treated soils compared to the control. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

Kuzyakov, 2013; Lehtoranta et al., 2015; Khan et al., 2019; Chen et al., 2020; Teng et al., 2020). Phosphorus in the soil solution (*e.g.*, HPO_4^{2-}, $H_2PO_4^-$) can be transformed into organic P through microbial assimilation and metabolism (Richardson and Simpson, 2011). The increase in some soil organic P fractions (*e.g.*, $NaHCO_3$-P_o, NaOH-P_o in bulk soil) after glucose addition supports the microbial immobilization pathway (Table 1). As plant growth requires phosphorus, we attribute the lack of an increase in available P in rhizosphere soil of the G5 treatment to uptake of P by the maize (Fig. 1a). However, P concentrations in maize aboveground biomass were not affected by glucose treatments (Fig. S1b). This indicates that glucose can stimulate plant growth through other mechanisms besides enhancing P uptake. In addition to their ability to solubilize P, phosphorus-solubilizing microbes may promote plant growth by asymbiotic N_2 fixation, plant hormones production, and control of phytopathogenic microorganisms (Rangarajan et al., 2003). For example, Pereira and Castro (2014) found that increased maize growth was not only related to P solubilization, but also to increased indol-3-acetic acid and ACC-deaminase activities. Furthermore, the enhanced microbial activity induced by C addition can affect several important soil processes, such as C and N cycling, thereby providing more nutrients for plant growth (Bornø et al., 2018). Hence, labile C additions mediated the transformation of soil P fractions from non-labile P pools to more labile P forms. This was consistent with the results from Sun et al. (2019) showing that cellulose promoted the transformation of citrate-P and HCl–P (insoluble P forms) to microbial P (labile P pool) via microbial enrichment in upland soils.

Compared with the control treatment (CK), we found a higher pH in glucose treated soils, both with and without maize (Fig. S1c). The priming effect induced by glucose addition may increase organic matter decomposition, with the decarboxylation of organic anions consuming protons and thereby increasing soil pH (Yan et al., 1996). Further, labile C provides energy and electron donors for the reduction of nitrate, and the decomposition of organic C consumes O_2 in the soil. Oxygen depletion may lead to anaerobic conditions (especially in microsites) that enhance dissimilatory reduction of nitrate (Fernandes et al., 2012; Lu et al., 2013). Hence, enhanced denitrification in glucose treated soils could also lead to an increase in soil pH. The forms and availability of soil P are strongly determined by soil pH and soil mineralogy (*e.g.*, Fe/Al minerals) (Hou et al., 2018). In acidic soils, elevated pH generally increases soil P availability through the release of phosphate from Fe/Al

minerals (Barrow, 1984; Goldberg and Sposito, 1984). This premise is consistent with the significant positive correlation between available P and soil pH in our study (Fig. S5).

Acid and alkaline phosphomonoesterase activities are commonly used as indicators of potential organic P mineralization (Chen, 2003). In soils, phosphodiesters can be hydrolyzed to monoesters by phosphodiesterase, which are subsequently transformed to plant-available P forms by acid and alkaline phosphomonoesterases (Tabatabai, 1994; Turner and Haygarth, 2005). For example, alanine amendment caused a 6-fold increase in phosphomonoesterase activity and a 31-fold increase in total P mineralization rate (Spohn et al., 2013). Similarly, ascorbic acid increased available P content by 1.9–202.7% through stimulating the activity of phosphomonoesterase in a paddy soil (Li et al., 2015). In this study, we found significant positive correlations between ALP activity and *phoD* gene abundance (Fig. S5) and between *phoD* gene abundance and the relative abundances of several specific *phoD* harboring bacterial and fungal genera (Fig. S4). These relationships suggest that glucose addition increased the microbial enzymatic activity for organic P mineralization through enhancing microbial growth. Further support derives from the significant increases in MBC and MBP concentrations (Fig. 2a and b). We found a higher microbial P/C ratio in glucose treated soils (Fig. 2c), that is attributed to P immobilization by microbes due to their enhanced activity. These microbes eventually replenish the soluble P pools via microbial turnover (Kwabiah et al., 2003). Therefore, microbial demand for carbon can facilitate the mineralization of organic P and increased levels of available P (Spohn et al., 2013; Heuck et al., 2015; Wang et al., 2016; Chen et al., 2020; Huang et al., 2021).

4.2. *phoD* harboring bacterial and fungal community response to labile C addition

Microorganisms need to assimilate P while mineralizing organic C to maintain a relative nutrient balance in microbial biomass (Cleveland and Liptzin, 2007; Heuck et al., 2015). Under P deficiency, phosphorus solubilizing microorganisms may enhance mineralization of organic P and solubilization of inorganic P to obtain the required P. We found significant variations in *phoD* harboring bacterial and fungal communities after glucose additions (Fig. S2). For *phoD* harboring bacteria, the relative abundance of *Bradyrhizobium* significantly increased in soil without maize and the G10-bulk soil (Fig. 3b and d). As previously demonstrated, *Bradyrhizobium* has the ability to promote the conversion of non-labile P to labile P forms through microbial immobilization and subsequent mineralization (Luo et al., 2017; Ibny et al., 2019; Zhu et al., 2021). Additionally, *Bradyrhizobium* harbors a N-fixing gene (Kaneko et al., 2002; Yao et al., 2014), suggesting that this genus could play an important role in coupled N/P cycling in soils with sufficient C. However, this was not observed in G10-rhizosphere soil (Fig. 3f). Root exudates (*e.g.*, amino acids, sugars, organic acid and protein) (Frey, 2007) may provide sufficient C to support rhizosphere microbial activities, thus exogenous C inputs may not stimulate the growth of phosphorus solubilizing microorganisms, such as *Bradyrhizobium*.

Within the fungal community, glucose additions significantly increased the relative abundance of *Eupenicillium* in all treated soils (Fig. 4). Labile C addition may promote the growth of specific fungal taxa that rapidly metabolize labile C substrates (Chigineva et al., 2009). *Eupenicillium* was previously reported to enhance phosphate solubilization in a range of soil environments (Vyas et al., 2007; Mendes et al., 2013, 2014). The positive correlation between *phoD* gene abundance and the relative abundance of *Eupenicillium* could infer a similar P solubilization role (Fig. S3). Given a greater supply of C substrates in the rhizosphere, fewer genera with decreased relative abundances were found in rhizosphere soil compared to bulk soils, and in soil without maize, after glucose additions (Fig. 4). Though the relative abundance of a few taxa changed in rhizosphere soil, we found enhanced rhizosphere microbial activity, which contributed to increased *phoD* gene abundance and ACP/ALP activities in response to glucose additions.

We found that soil pH, *phoD* gene abundance and potential ACP activity were the main factors accounting for the variation of *phoD* harboring bacterial and fungal community structure (Fig. S4). Since ALP is derived from the expression of the *phoD* gene, bacterial community structure appears to be related to *phoD* gene abundance rather than ALP activity. Soil pH is generally found to be a key factor affecting bacterial community structure (Lauber et al., 2008; Griffiths et al., 2011), whereas the fungal community is relatively stable within the pH range from 3.6 to 6.0 (Lauber et al., 2008; Ragot et al., 2016). The *phoD* gene, which encodes alkaline phosphatase, is widespread across the bacterial kingdom, but is also found in archaea and fungi (Ragot et al., 2015). Therefore, the increase of *phoD* gene abundance in glucose treated soils (Fig. 2d) was ascribed to the alteration of the *phoD* harboring bacterial community. Both bacteria and fungi have the ability to secrete acid phosphatase (Nannipieri et al., 2011). Our results were in agreement with previous studies that found ACP activity was increased by microorganisms experiencing P deficiency (Krey et al., 2011; Ragot et al., 2016, 2017). As plants are also able to produce acid phosphatase (Tadano and Sakai, 1991), ACP activity in soils with maize may further originate from plant roots.

4.3. Labile C regulates P availability via microbial interactions

Generally, soil microorganisms do not live independently, instead they form complex inter-species networks within ecological communities that subsequently regulate ecosystem functions (Fuhrman, 2009; Freilich et al., 2010). In the networks of *phoD* harboring bacteria, glucose additions enhanced the interactions (*i.e.*, links) between phosphorus solubilizing bacteria, particularly increasing the connections between *Bradyrhizobium* and other genera (Fig. 5). Thus, the *phoD* harboring bacterial communities in the G5 and G10 treatments formed more complex networks, suggesting that glucose additions may improve soil P availability through more cooperation/interactions between phosphorus solubilizing bacteria. In contrast to the *phoD* harboring bacterial networks, glucose additions decreased the connections between fungal genera and displayed a weaker network complexity compared with the control (Fig. 5). This may result from competition between soil bacteria and fungi for C-substrates (DeCrappeo et al., 2017; Hicks et al., 2019). Fungal OTUs were predominant in the combined *phoD* harboring bacterial and fungal networks of the CK treatment, whereas *phoD* harboring bacterial OTUs dominated the networks of the G5 and G10 treatments (Table S2b). Thus, in both individual and combined networks, we observed that glucose additions enhanced interactions between *phoD* harboring bacteria relative to that with fungi. Bacteria can respond quickly to increased concentrations of labile C-substrates (Paterson et al., 2008; Dungait et al., 2013), and hence dominated the mineralization of added glucose in our study. The classic food web model also assumes that increased labile C preferentially facilitates the proliferation of bacteria (de Vries and Caruso, 2016). In sum, our results indicated that the positive effect of labile C inputs on P availability in a P-deficient soil was mediated by synergism between bacteria and fungi, but bacterial interactions dominated the release of P.

5. Conclusions

Two dose rates of glucose (5 and 10 mg glucose-C g^{-1} soil) increased available P concentrations in soils without maize and in the bulk soil with maize, as well as in the rhizosphere soil receiving 10 mg glucose-C g^{-1} soil. The enhanced P availability increased aboveground/belowground biomass and P uptake by maize. We observed significant positive correlations of available P with *phoD* gene abundance, potential ACP/ALP activities and soil pH. The altered *phoD* harboring bacterial and fungal community structure contributed to increased potential ACP activities and *phoD* gene abundance, and further caused enhanced potential ALP activities in glucose treated soils. Our study provides evidence that labile C can drive the transformation of non-available P to available

P forms in a P-deficient soil through enhancing the growth of specific bacterial and fungal taxa (*e.g.*, *Bradyrhizobium* and *Eupenicillium*) and promoting interactions between P solubilizing bacteria in the maize rhizosphere. Future studies may garner additional information from more intensive temporal sampling and investigation of a gradient of soils with contrasting P status to further clarify the universality of labile C input on soil P availability and plant uptake.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

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References

Abbruzzese, V., Semple, K.T., Haygarth, P.M., Aller, M.F., Russell, E., Surridge, B.W.J., 2021. Effects of substrate quality on carbon partitioning and microbial community composition in soil from an agricultural grassland. Applied Soil Ecology 161, 103881.

Barrow, N.J., 1984. Modeling the effects of pH on phosphate sorption by soils. Journal of Soil Science 35, 283–297.

Bornø, M.L., Eduah, J.O., Müller-Stöver, D.S., Liu, F.L., 2018. Effect of different biochars on phosphorus (P) dynamics in the rhizosphere of *Zea mays* L. (maize). Plant and Soil 431, 257–272.

Brookes, P.C., Powlson, D.S., Jenkinson, D.S., 1982. Measurement of microbial biomass phosphorus in soil. Soil Biology and Biochemistry 14, 319–329.

Chatterjee, D., Nayak, A.K., Mishra, A., Swain, C.K., Kumar, U., Bhaduri, D., Panneerselvam, P., Lal, B., Gautam, P., Pathak, H., 2021. Effect of long-term organic fertilization in flooded rice soil on phosphorus transformation and phosphate solubilizing microorganisms. Journal of Soil Science and Plant Nutrition 21, 1368–1381.

Chen, H.J., 2003. Phosphatase activity and P fractions in soils of an 18-year-old Chinese fir (*Cunninghamia lanceolata*) plantation. Forest Ecology and Management 178, 301–310.

Chen, Y., Sun, R.B., Sun, T.T., Chen, P., Yu, Z.Y., Ding, L.Y., Jiang, Y.J., Wang, X.Y., Dai, C.C., Sun, B., 2020. Evidence for involvement of keystone fungal taxa in organic phosphorus mineralization in subtropical soil and the impact of labile carbon. Soil Biology and Biochemistry 148, 107900.

Chigineva, N.I., Aleksandrova, A.V., Tiunov, A.V., 2009. The addition of labile carbon alters litter fungal communities and decreases litter decomposition rates. Applied Soil Ecology 42, 264–270.

Cleveland, C.C., Liptzin, D., 2007. C:N:P stoichiometry in soil: is there a "Redfield ratio" for the microbial biomass? Biogeochemistry 85, 235–252.

Dai, Z.M., Liu, G.F., Chen, H.H., Chen, C.R., Wang, J.K., Ai, S.Y., Wei, D., Li, D.M., Ma, B., Tang, C.X., Brookes, P.C., Xu, J.M., 2020. Long-term nutrient inputs shift soil microbial functional profiles of phosphorus cycling in diverse agroecosystems. The ISME Journal 14, 757–770.

Dai, Z.M., Su, W.Q., Chen, H.H., Barberan, A., Zhao, H.C., Yu, M.J., Yu, L., Brookes, P.C., Schadt, C.W., Chang, S.X., Xu, J.M., 2018. Long-term nitrogen fertilization decreases bacterial diversity and favors the growth of *Actinobacteria* and *Proteobacteria* in agro-ecosystems across the globe. Global Change Biology 24, 3452–3461.

de Vries, F.T., Caruso, T., 2016. Eating from the same plate? Revisiting the role of labile carbon inputs in the soil food web. Soil Biology and Biochemistry 102, 4–9.

DeCrappeo, N.M., DeLorenze, E.J., Giguere, A.T., Pyke, D.A., Bottomley, P.J., 2017. Fungal and bacterial contributions to nitrogen cycling in cheatgrass-invaded and uninvaded native sagebrush soils of the western USA. Plant and Soil 416, 271–281.

Ding, L.J., Su, J.Q., Sun, G.X., Wu, J.S., Wei, W.X., 2018. Increased microbial functional diversity under long-term organic and integrated fertilization in a paddy soil. Applied Microbiology and Biotechnology 102, 1969–1982.

Dungait, J.A.J., Kemmitt, S.J., Michallon, L., Guo, S., Wen, Q., Brookes, P.C., Evershed, R.P., 2013. The variable response of soil microorganisms to trace concentrations of low molecular weight organic substrates of increasing complexity. Soil Biology and Biochemistry 64, 57–64.

Edgar, R.C., 2010. Search and clustering orders of magnitude faster than BLAST. Bioinformatics 26, 2460–2461.

Edgar, R.C., 2013. UPARSE: highly accurate OTU sequences from microbial amplicon reads. Nature Methods 10, 996–998.

Fabian, J., Zlatanovic, S., Mutz, M., Premke, K., 2017. Fungal-bacterial dynamics and their contribution to terrigenous carbon turnover in relation to organic matter quality. The ISME Journal 11, 415–425.

Fernandes, S.O., Bonin, P.C., Michotey, V.D., Garcia, N., LokaBharathi, P.A., 2012. Nitrogen-limited mangrove ecosystems conserve N through dissimilatory nitrate reduction to ammonium. Scientific Reports 2, 419.

Fierer, N., 2017. Embracing the unknown: disentangling the complexities of the soil microbiome. Nature Reviews Microbiology 15, 579–590.

Fraser, T., Lynch, D.H., Entz, M.H., Dunfield, K.E., 2015. Linking alkaline phosphatase activity with bacterial *phoD* gene abundance in soil from a long-term management trial. Geoderma 257, 115–122.

Freilich, S., Kreimer, A., Meilijson, I., Gophna, U., Sharan, R., Ruppin, E., 2010. The large-scale organization of the bacterial network of ecological co-occurrence interactions. Nucleic Acids Research 38, 3857–3868.

Frey, S.D., 2007. Spatial distribution of soil organisms. In: Paul, E.A. (Ed.), Soil Microbiology, Ecology, and Biochemistry, third ed. Elsevier Inc., USA, Burlington, MA, pp. 283–300.

Fuhrman, J.A., 2009. Microbial community structure and its functional implications. Nature 459, 193–199.

George, T.S., Giles, C.D., Menezes-Blackburn, D., Condron, L.M., Gama-Rodrigues, A.C., Jaisi, D., Lang, F., Neal, A.L., Stutter, M.I., Almeida, D.S., Bol, R., Cabugao, K.G., Celi, L., Cotner, J.B., Feng, G., Goll, D.S., Hallama, M., Krueger, J., Plassard, C., Rosling, A., Darch, T., Fraser, T., Giesler, R., Richardson, A.E., Tamburini, F., Shand, C.A., Lumsdon, D.G., Zhang, H., Blackwell, M.S.A., Wearing, C., Mezeli, M. M., Almas, A.R., Audette, Y., Bertrand, I., Beyhaut, E., Boitt, G., Bradshaw, N., Brearley, C.A., Bruulsema, T.W., Ciais, P., Cozzolino, V., Duran, P.C., Mora, M.L., de Menezes, A.B., Dodd, R.J., Dunfield, K., Engl, C., Frazao, J.J., Garland, G., Jimenez, J.L.G., Graca, J., Granger, S.J., Harrison, A.F., Heuck, C., Hou, E.Q., Johnes, P.J., Kaiser, K., Kjaer, H.A., Klumpp, E., Lamb, A.L., Macintosh, K.A., Mackay, E.B., McGrath, J., McIntyre, C., McLaren, T., Meszaros, E., Missong, A., Mooshammer, M., Negron, C.P., Nelson, L.A., Pfahler, V., Poblete-Grant, P., Randall, M., Seguel, A., Seth, K., Smith, A.C., Smits, M.M., Sobarzo, J.A., Spohn, M., Tawaraya, K., Tibbett, M., Voroney, P., Wallander, H., Wang, L., Wasaki, J., Haygarth, P.M., 2018. Organic phosphorus in the terrestrial environment: a perspective on the state of the art and future priorities. Plant and Soil 427, 191–208.

Gerke, J., 2015. The acquisition of phosphate by higher plants: effect of carboxylate release by the roots. A critical review. Journal of Plant Nutrition and Soil Science 178, 351–364.

Goldberg, S., Sposito, G., 1984. A chemical-model of phosphate adsorption by soils: II. Noncalcareous soils. Soil Science Society of America Journal 48, 779–783.

Griffiths, R.I., Thomson, B.C., James, P., Bell, T., Bailey, M., Whiteley, A.S., 2011. The bacterial biogeography of British soils. Environmental Microbiology 13, 1642–1654.

He, L.Z., Dai, Z.M., Liu, X.M., Tang, C.X., Xu, J.M., 2021. Effect of alkaline lignin on immobilization of cadmium and lead in soils and the associated mechanisms. Chemosphere 281, 130969.

Hedley, M.J., Stewart, J.W.B., Chauhan, B.S., 1982. Changes in inorganic and organic soil-phosphorus fractions induced by cultivation practices and by laboratory incubations. Soil Science Society of America Journal 46, 970–976.

Heuck, C., Weig, A., Spohn, M., 2015. Soil microbial biomass C:N:P stoichiometry and microbial use of organic phosphorus. Soil Biology and Biochemistry 85, 119–129.

Hicks, L.C., Ang, R., Leizeaga, A., Rousk, J., 2019. Bacteria constrain the fungal growth response to drying-rewetting. Soil Biology and Biochemistry 134, 108–112.

Hou, E.Q., Wen, D.Z., Kuang, Y.W., Cong, J., Chen, C.R., He, X.J., Heenan, M., Lu, H., Zhang, Y.G., 2018. Soil pH predominantly controls the forms of organic phosphorus in topsoils under natural broadleaved forests along a 2500 km latitudinal gradient. Geoderma 315, 65–74.

Huang, Y.L., Dai, Z.M., Lin, J.H., Qi, Q., Luo, Y., Dahlgren, R.A., Xu, J.M., 2021. Contrasting effects of carbon source recalcitrance on soil phosphorus availability and communities of phosphorus solubilizing microorganisms. Journal of Environmental Management 298, 113426.

Ibny, F.Y.I., Jaiswal, S.K., Mohammed, M., Dakora, F.D., 2019. Symbiotic effectiveness and ecologically adaptive traits of native rhizobial symbionts of Bambara groundnut (*Vigna subterranea* L. Verdc.) in Africa and their relationship with phylogeny. Scientific Reports 9, 12666.

Kaneko, T., Nakamura, Y., Sato, S., Minamisawa, K., Uchiumi, T., Sasamoto, S., Watanabe, A., Idesawa, K., Iriguchi, M., Kawashima, K., Kohara, M., Matsumoto, M., Shimpo, S., Tsuruoka, H., Wada, T., Yamada, M., Tabata, S., 2002. Complete genomic sequence of nitrogen-fixing symbiotic bacterium *Bradyrhizobium japonicum* USDA110. DNA Research 9, 189–197.

Kathuria, S., Martiny, A.C., 2011. Prevalence of a calcium-based alkaline phosphatase associated with the marine cyanobacterium *Prochlorococcus* and other ocean bacteria. Environmental Microbiology 13, 74–83.

Khan, I., Fahad, S., Wu, L., Zhou, W., Xu, P., Sun, Z., Salam, A., Imran, M., Jiang, M.D., Kuzyakov, Y., Hu, R.G., 2019. Labile organic matter intensifies phosphorous mobilization in paddy soils by microbial iron (III) reduction. Geoderma 352, 185–196.

Kishore, N., Pindi, P.K., Ram-Reddy, S., 2015. Phosphate-solubilizing microorganisms: a critical review. In: Bahadur, B., Venkat-Rajam, M., Sahijram, L., Krishnamurthy, K. (Eds.), Plant Biology and Biotechnology. Springer, New Delhi, pp. 307–333.

Kochian, L.V., 2012. Plant nutrition rooting for more phosphorus. Nature 488, 466–467.

Krauss, G.J., Sole, M., Krauss, G., Schlosser, D., Wesenberg, D., Baerlocher, F., 2011. Fungi in freshwaters: ecology, physiology and biochemical potential. FEMS Microbiology Reviews 35, 620–651.
Krey, T., Caus, M., Baum, C., Ruppel, S., Eichler-Loebermann, B., 2011. Interactive effects of plant growth-promoting rhizobacteria and organic fertilization on P nutrition of *Zea mays* L. and *Brassica napus* L. Journal of Plant Nutrition and Soil Science 174, 602–613.
Kwabiah, A.B., Palm, C.A., Stoskopf, N.C., Voroney, R.P., 2003. Response of soil microbial biomass dynamics to quality of plant materials with emphasis on P availability. Soil Biology and Biochemistry 35, 207–216.
Lauber, C.L., Strickland, M.S., Bradford, M.A., Fierer, N., 2008. The influence of soil properties on the structure of bacterial and fungal communities across land-use types. Soil Biology and Biochemistry 40, 2407–2415.
Lehtoranta, J., Ekholm, P., Wahlström, S., Tallberg, P., Uusitalo, R., 2015. Labile organic carbon regulates phosphorus release from eroded soil transported into anaerobic coastal systems. Ambio 44, 263–273.
Li, X.L., Luo, L.J., Yang, J.S., Li, B.Z., Yuan, H.L., 2015. Mechanisms for solubilization of various insoluble phosphates and activation of immobilized phosphates in different soils by an efficient and salinity-tolerant *Aspergillus niger* Strain An2. Applied Biochemistry and Biotechnology 175, 2755–2768.
Lu, W.W., Zhang, H.L., Shi, W.M., 2013. Dissimilatory nitrate reduction to ammonium in an anaerobic agricultural soil as affected by glucose and free sulfide. European Journal of Soil Biology 58, 98–104.
Luo, G.W., Ling, N., Nannipieri, P., Chen, H., Raza, W., Wang, M., Guo, S.W., Shen, Q.R., 2017. Long-term fertilisation regimes affect the composition of the alkaline phosphomonoesterase encoding microbial community of a Vertisol and its derivative soil fractions. Biology and Fertility of Soils 53, 375–388.
Ma, B., Stirling, E., Liu, Y.H., Zhao, K.K., Zhou, J.Z., Singh, B.K., Tang, C.X., Dahlgren, R. A., Xu, J.M., 2021. Soil biogeochemical cycle couplings inferred from a function-taxon network. Research 2021, 7102769.
Mendes, G.O., Dias, C.S., Silva, I.R., Ribeiro-Junior, J.I., Pereira, O.L., Costa, M.D., 2013. Fungal rock phosphate solubilization using sugarcane bagasse. World Journal of Microbiology and Biotechnology 29, 43–50.
Mendes, G.O., Moreira de Freitas, A.L., Pereira, O.L., da Silva, I.R., Bojkov-Vassilev, N., Costa, M.D., 2014. Mechanisms of phosphate solubilization by fungal isolates when exposed to different P sources. Annals of Microbiology 64, 239–249.
Munkvold, L., Kjoller, R., Vestberg, M., Rosendahl, S., Jakobsen, I., 2004. High functional diversity within species of arbuscular mycorrhizal fungi. New Phytologist 164, 357–364.
Murphy, J., Riley, J.P., 1962. A modified single solution method for determination of phosphate in natural waters. Analytica Chimica Acta 26, 31–36.
Nannipieri, P., Giagnoni, L., Landi, L., Renella, G., 2011. Role of phosphatase enzymes in soil. In: Bünemann, E.K., Oberson, A., Frossard, E. (Eds.), Phosphorus in Action. Springer, Berlin, pp. 215–243.
Parks, D.H., Tyson, G.W., Hugenholtz, P., Beiko, R.G., 2014. STAMP: statistical analysis of taxonomic and functional profiles. Bioinformatics 30, 3123–3124.
Paterson, E., Osler, G., Dawson, L.A., Gebbing, T., Sim, A., Ord, B., 2008. Labile and recalcitrant plant fractions are utilised by distinct microbial communities in soil: independent of the presence of roots and mycorrhizal fungi. Soil Biology and Biochemistry 40, 1103–1113.
Pereira, S.I.A., Castro, P.M.L., 2014. Phosphate-solubilizing rhizobacteria enhance *Zea mays* growth in agricultural P-deficient soils. Ecological Engineering 73, 526–535.
Qaswar, M., Ahmed, W., Huang, J., Fan, H.Z., Shi, X.J., Jiang, X.J., Liu, K.L., Xu, Y.M., He, Z.Q., Asghar, W., Shah, A., Zhang, H.M., 2019. Soil carbon (C), nitrogen (N) and phosphorus (P) stoichiometry drives phosphorus lability in paddy soil under long-term fertilization: a fractionation and path analysis study. PLoS One 14, e0218195.
Ragot, S.A., Huguenin-Elie, O., Kertesz, M.A., Frossard, E., Bünemann, E.K., 2016. Total and active microbial communities and *phoD* as affected by phosphate depletion and pH in soil. Plant and Soil 408, 15–30.
Ragot, S.A., Kertesz, M.A., Bünemann, E.K., 2015. *phoD* alkaline phosphatase gene diversity in soil. Applied and Environmental Microbiology 81, 7281–7289.
Ragot, S.A., Kertesz, M.A., Meszaros, E., Frossard, E., Bunemann, E.K., 2017. Soil *phoD* and *phoX* alkaline phosphatase gene diversity responds to multiple environmental factors. FEMS Microbiology Ecology 93, fiw212.
Rangarajan, S., Saleena, L.M., Vasudevan, P., Nair, S., 2003. Biological suppression of rice disease by *Pseudomonas* spp. under saline soil condition. Plant and Soil 251, 73–82.
Richardson, A.E., 2001. Prospects for using soil microorganisms to improve the acquisition of phosphorus by plants. Australian Journal of Plant Physiology 28, 897–906.
Richardson, A.E., Simpson, R.J., 2011. Soil microorganisms mediating phosphorus availability. Plant Physiology 156, 989–996.
Rognes, T., Flouri, T., Nichols, B., Quince, C., Mahe, F., 2016. VSEARCH: a versatile open source tool for metagenomics. PeerJ 4, e2584.
Sakurai, M., Wasaki, J., Tomizawa, Y., Shinano, T., Osaki, M., 2008. Analysis of bacterial communities on alkaline phosphatase genes in soil supplied with organic matter. Soil Science and Plant Nutrition 54, 62–71.
Sharma, S.B., Sayyed, R.Z., Trivedi, M.H., Gobi, T.A., 2013. Phosphate solubilizing microbes: sustainable approach for managing phosphorus deficiency in agricultural soils. SpringerPlus 2, 587.
Smith, S.E., Smith, F.A., 2011. Roles of arbuscular mycorrhizas in plant nutrition and growth: new paradigms from cellular to ecosystem scales. Annual Review of Plant Biology 62, 227–250.
Spohn, M., Ermak, A., Kuzyakov, Y., 2013. Microbial gross organic phosphorus mineralization can be stimulated by root exudates–a ^{33}P isotopic dilution study. Soil Biology and Biochemistry 65, 254–263.
Spohn, M., Kuzyakov, Y., 2013. Phosphorus mineralization can be driven by microbial need for carbon. Soil Biology and Biochemistry 61, 69–75.
Sun, Q., Qiu, H.S., Hu, Y.J., Wei, X.M., Chen, X.B., Ge, T.D., Wu, J.S., Su, Y.R., 2019. Cellulose and lignin regulate partitioning of soil phosphorus fractions and alkaline phosphomonoesterase encoding bacterial community in phosphorus-deficient soils. Biology and Fertility of Soils 55, 31–42.
Tabatabai, M.A., 1994. Soil enzymes. In: Weaver, R.W., Angle, S., Bottomley, P., Bezdicek, D., Smith, S., Tabatabai, A., Wollum, A. (Eds.), Methods of Soil Analysis: Part 2. Microbiological and Biochemical Properties. Soil Science Society of America, Madison, pp. 775–833.
Tadano, T., Sakai, H., 1991. Secretion of acid-phosphatase by the roots of several crop species under phosphorus-deficient conditions. Soil Science and Plant Nutrition 37, 129–140.
Tan, H., Barret, M., Mooij, M.J., Rice, O., Morrissey, J.P., Dobson, A., Griffiths, B., O'Gara, F., 2013. Long-term phosphorus fertilisation increased the diversity of the total bacterial community and the *phoD* phosphorus mineraliser group in pasture soils. Biology and Fertility of Soils 49, 661–672.
Tedersoo, L., Bahram, M., Polme, S., Koljalg, U., Yorou, N.S., Wijesundera, R., Villarreal-Ruiz, L., Vasco-Palacios, A.M., Pham-Quang, T., Suija, A., Smith, M.E., Sharp, C., Saluveer, E., Saitta, A., Rosas, M., Riit, T., Ratkowsky, D., Pritsch, K., Poldmaa, K., Piepenbring, M., Phosri, C., Peterson, M., Parts, K., Paertel, K., Otsing, E., Nouhra, E., Njouonkou, A.L., Nilsson, R.H., Morgado, L.N., Mayor, J., May, T.W., Majuakim, L., Lodge, D.J., Lee, S.S., Larsson, K.H., Kohout, P., Hosaka, K., Hiiesalu, I., Henkel, T. W., Harend, H., Guo, L.D., Greslebin, A., Grelet, G., Geml, J., Gates, G., Dunstan, W., Dunk, C., Drenkhan, R., Dearnaley, J., De Kesel, A., Tan, D., Chen, X., Buegger, F., Brearley, F.Q., Bonito, G., Anslan, S., Abell, S., Abarenkov, K., 2014. Global diversity and geography of soil fungi. Science 346, 1256688.
Teng, Z.D., Zhu, J., Shao, W., Zhang, K.Y., Li, M., Whelan, M.J., 2020. Increasing plant availability of legacy phosphorus in calcareous soils using some phosphorus activators. Journal of Environmental Management 256, 109952.
Tiessen, H., Moir, J.O., 1993. Characterization of available P by sequential fractionation. In: Carter, M.R. (Ed.), Soil Sampling and Methods of Analysis. Canadian Society of Soil Science, Lewis Publishers, Boca Raton, FL, pp. 75–86.
Turner, B.L., Haygarth, P.M., 2005. Phosphatase activity in temperate pasture soils: potential regulation of labile organic phosphorus turnover by phosphodiesterase activity. Science of the Total Environment 344, 27–36.
Václavková, Š., Šyc, M., Moško, J., Pohořelý, M., Svoboda, K., 2018. Fertilizer and soil solubility of secondary P sources–the estimation of their applicability to agricultural soils. Environmental Science and Technology 52, 9810–9817.
van der Heijden, M.G.A., Bardgett, R.D., van Straalen, N.M., 2008. The unseen majority: soil microbes as drivers of plant diversity and productivity in terrestrial ecosystems. Ecology Letters 11, 296–310.
van der Heijden, M.G.A., Streitwolf-Engel, R., Riedl, R., Siegrist, S., Neudecker, A., Ineichen, K., Boller, T., Wiemken, A., Sanders, I.R., 2006. The mycorrhizal contribution to plant productivity, plant nutrition and soil structure in experimental grassland. New Phytologist 172, 739–752.
Vyas, P., Rahi, P., Chauhan, A., Gulati, A., 2007. Phosphate solubilization potential and stress tolerance of *Eupenicillium parvum* from tea soil. Mycological Research 111, 931–938.
Wang, J.P., Wu, Y.H., Zhou, J., Bing, H.J., Sun, H.Y., 2016. Carbon demand drives microbial mineralization of organic phosphorus during the early stage of soil development. Biology and Fertility of Soils 52, 825–839.
Wei, X.M., Hu, Y.J., Razavi, B.S., Zhou, J., Shen, J.L., Nannipieri, P., Wu, J.S., Ge, T.D., 2019. Rare taxa of alkaline phosphomonoesterase-harboring microorganisms mediate soil phosphorus mineralization. Soil Biology and Biochemistry 131, 62–70.
White, T., Bruns, T., Lee, S., Taylor, J., 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis, N., Gelfand, D., Sninsky, J., White, T. (Eds.), PCR: Protocols and Applications–A Laboratory Manual. Academic Press, New York, pp. 315–322.
Yan, F., Schubert, S., Mengel, K., 1996. Soil pH increase due to biological decarboxylation of organic anions. Soil Biology and Biochemistry 28, 617–624.
Yao, Y., Wang, R., Lu, J.K., Sui, X.H., Wang, E.T., Chen, W.X., 2014. Genetic diversity and evolution of *Bradyrhizobium* populations nodulating *Erythrophleum fordii*, an evergreen tree indigenous to the southern subtropical region of China. Applied and Environmental Microbiology 80, 6184–6194.
Zhu, X.Y., Zhao, X.R., Lin, Q.M., Li, G.T., 2021. Distribution characteristics of *phoD*-harbouring bacterial community structure and its roles in phosphorus transformation in steppe soils in northern China. Journal of Soil Science and Plant Nutrition 21, 1531–1541.