

Crop acquisition of phosphorus, iron and zinc from soil in cereal/legume intercropping systems: A critical review

Yanfang Xue¹, Haiyong Xia^{1,2,*}, Peter Christie³, Zheng Zhang², Long Li³ and Caixian Tang^{4,*}

¹*National Engineering Laboratory for Wheat and Maize, Maize Research Institute, Shandong Academy of Agricultural Sciences, Jinan 250100, China,*

²*Shandong Provincial Key Laboratory of Crop Genetic Improvement, Ecology and Physiology, Crop Research Institute, Shandong Academy of Agricultural Sciences, Jinan 250100, China,*

³*Ministry of Education Key Laboratory of Plant and Soil Interactions, Center for Resources, Environment and Food Security, China Agricultural University, Beijing 100193, China and*

⁴*Department of Animal, Plant and Soil Sciences, Centre for AgriBiosciences, La Trobe University, Melbourne Campus, Bundoora Vic. 3086, Australia*

Abstract

● *Background* Phosphorus (P), iron (Fe) and zinc (Zn) are essential elements for plant growth and development but their availability in soil is often limited. Intercropping contributes to increased P, Fe and Zn uptake and thereby increases yield and improves grain nutritional quality and ultimately human health. A better understanding how intercropping leads to increased plant P, Fe and Zn availability helps to improve P-fertilizer-use efficiency and agronomic Fe and Zn biofortification.

● *Scope* This review synthesizes the literature on how intercropping of legumes with cereals increases acquisition of P, Fe and Zn from soil and recapitulates what is known about root-to-shoot nutrient translocation, plant-internal nutrient remobilization and allocation to grains.

● *Conclusions* Direct interspecific facilitation in intercropping involves below-ground processes in which cereals increase Fe and Zn bioavailability while companion legumes benefit. This has been demonstrated and verified using isotopic nutrient tracing and molecular analysis. The same methodological approaches and field studies should be used to explore direct interspecific P facilitation. Both niche complementarity and interspecific facilitation contribute to increased P acquisition in intercropping. Niche complementarity may also contribute to increased Fe and Zn acquisition, an aspect poorly understood. Interspecific mobilization and uptake facilitation of sparingly soluble P, Fe and Zn from soil, however, do not solely determine concentrations of P, Fe and Zn in grains. Grain yield and nutrient translocation from roots to shoots further influence the concentrations of these nutrients in grains.

Key words: Biofortification, cereals, intercropping, interspecific facilitation, legumes, micronutrients, niche complementarity, phosphorus, rhizosphere, root architecture, root exudates, soil microbes.

INTRODUCTION

Agricultural phosphorus (P) inputs have greatly increased soil fertility and crop production worldwide. However, excessive applications of P fertilizers and P-rich manures have led to an accumulation of P in many soils, which lowers P-fertilizer-use efficiency and leads to P losses via runoff and eutrophication of surface waters (Zhang *et al.*, 2013). It is therefore important to enhance P-use efficiency in crop production through optimized application of P fertilizers and utilization of residual P and other P pools from soils. Root-microbe-soil interactions may play a key role in the effective use of P in this respect (Shen *et al.*, 2013).

Deficiencies of iron (Fe) and zinc (Zn) in food are a global problem, impairing human health in general and development of children and pregnant and lactating women in particular (Stein, 2010). Micronutrient biofortification, the process of enriching micronutrients including Fe and Zn in the edible parts of crops, represents a sustainable, economical and easily implementable solution to overcome micronutrient malnutrition (Bevis, 2015). Biofortification can be achieved through management of crops and soils, and by plant breeding and genetic transformation. Recent studies on micronutrient biofortification have focused on management practices in single-species cropping (Xue *et al.*, 2012, 2014a, b, c; Soltani *et al.*, 2014), but less work has been done in intercropping systems.

Intercropping systems play an important role in increasing crop yield and quality and environmental quality through efficient utilization of land, light, water and soil nutrients (Brooker *et al.*, 2015). It is defined as growing individuals of at least two crop species in close proximity at (about) the same time, leading to enhanced interspecific interactions and crop diversity (L. Li *et al.*, 2014). The mechanisms underlying overall yield increase (over-yielding) of intercropping compared to monocropping systems have been explained by two major ecological principles leading to improved resource use: niche complementarity and interspecific facilitation (Fridley, 2001; Hauggaard-Nielsen and Jensen, 2005; Li *et al.*, 2007). The niche complementarity hypothesis suggests reduced competition due to complementary use of resources and niche differentiation between intercropped species in time, in space or in forms of a given resource (Tilman *et al.*, 2001).

Interspecific facilitation, as defined by Callaway (1995), is a beneficial effect of the individuals of one species on the growth or survival of the individuals of another species. This can occur directly through positive mechanisms, e.g. the amelioration of harsh environmental conditions, including favorable alteration of light, temperature and soil moisture and increased availability of a limiting soil nutrient (Sekiya *et al.*, 2011; L. Li *et al.*, 2014). Alternatively, facilitation may occur indirectly, e.g. via stimulation of beneficial soil microbes to release nutrients during mineralization of organic matter, mobilization of sparingly soluble inorganic nutrients, or transfer of nutrients through common mycorrhizal networks linking co-cultivated crop plant species (Bainard *et al.*, 2012; Walder *et al.*, 2012; Ehrmann and Ritz, 2013; Sun *et al.*, 2013; Tong *et al.*, 2015).

Most studies of nutrient efficiency in cereal/legume intercropping systems are concerned about nitrogen (N), especially with regard to legume N₂ fixation. Both hypotheses of ‘niche complementarity’ and ‘interspecific facilitation’ have been used mainly to explain such N use in legume/cereal intercropping systems (Hinsinger *et al.*, 2011). Studies on soil P or micronutrients such as Fe and Zn are relatively rare. Recently, this research field has received

more attention with the reported phenomena of enhanced soil P acquisition by cereals intercropped with legumes (Li *et al.*, 2007) and enhanced Fe and Zn uptake by legume crops intercropped with cereals (Zuo and Zhang, 2009), especially in China. Will niche complementarity and interspecific facilitation mechanisms in intercropping apply to crop uptake of P, Fe and Zn from sparingly soluble sources in soils? Some experiments have been performed on P and on Fe/Zn, respectively, in order to answer this question. Improved P, Fe and Zn uptake and utilization in intercropping are expected to influence grain nutritional quality (Dey, 2003; Musa *et al.*, 2011; Briat *et al.*, 2015; Dragicevic *et al.*, 2015). However, the effects of intercropping on the uptake of P, Fe and Zn by crops and accumulation in grains are largely unknown.

This paper reviews and synthesizes the literature on how intercropping of legumes and cereals influences the mobilization and acquisition of P, Fe and Zn from soils, and recapitulates what is known about root-to-shoot nutrient translocation, plant internal nutrient remobilization and allocation to grains. It also identifies topics in need of further research.

PHOSPHORUS ACQUISITION FROM SOIL – ADVANTAGES AND MECHANISMS

Observations

Intercropping has been shown to increase the acquisition of P from sparingly soluble P in soils or other culture substrates in a wide range of systems including pot, rhizobox and hydroponic experiments, and under field conditions (Table 1). Intercropping of cereal/legume crops is the most commonly used system reported in the literature. Most studies have shown that intercropping increases P acquisition from organic or inorganic P sources.

There are three possible outcomes of increased P acquisition in the whole intercropping system compared to the monocropping system (Table 1). First, P acquisition and yields of both crop species are enhanced. For example, maize/faba bean intercropping increased P acquisition and the grain yield per unit area of the maize row by 23.9% and 49.0%, respectively, and of the faba bean row by 37.3% and 22.0%, respectively, in P-deficient calcareous soils over four years in a field experiment (Li *et al.*, 2007). Similar results were found by Mei *et al.* (2012) in a reclaimed desert soil. The second outcome of intercropping is an increase in P acquisition of one crop species and no change in the other crop. For example, in experiments of wheat/lupin in low-P sandy soils (Gardner and Boundy, 1983; Cu *et al.*, 2005) and maize/faba bean in a calcareous soil (H. Li *et al.*, 2014) intercropping, P acquisition of wheat or faba bean on a per-plant basis increased without influencing that of the companion crop. However, for the second outcome, the yield situation of each crop species in intercropping versus monocropping may vary under different conditions. The third outcome is that a crop species may increase the P uptake and yield of an accompanying crop species to its own detriment (Li *et al.*, 2003a; S. Li *et al.*, 2004; Hauggaard-Nielsen *et al.*, 2009; Xia *et al.*, 2013a), as observed in a maize/soybean intercropping in a calcareous soil (Xia *et al.*, 2013a). Maize experienced an intercropping advantage in P acquisition and grain yield per unit area of the maize row but soybean, although mobilizing P, exhibited a decline in P uptake and grain yield.

Complementary P acquisition from soil P pools

Both spatial and temporal niche complementarities in root distribution are important in

determining the amount of P accessible to plants because of the low mobility of P in soils. Spatial complementarity can occur between two crop species with contrasting root architecture, leading to exploitation of different soil horizons (Li *et al.*, 2006; C.C. Zhang *et al.*, 2014; Fig. 1). For example, root compatibility between intercropped maize and associated faba bean allowed intercropped maize to spread underneath the roots of neighboring faba bean, increasing the soil space exploited for nutrients and water (Li *et al.*, 2006). Intercropped species with contrasting phenologies (Rose *et al.*, 2007) and/or growth periods (e.g. different sowing/harvest dates) (Li *et al.*, 1999, 2007) may show differential requirements of nutrients (e.g. P and N) over time (Dessougi *et al.*, 2003; Bainard *et al.*, 2012; Amossé *et al.*, 2014). Such temporal niche complementarity (Fig. 1) also allows for the later crop to receive mineral nutrients from mineralization of residues of an earlier harvested companion crop (Hinsinger *et al.*, 2011). Relay intercropping, in which a second crop is planted before the first crop matures to grow simultaneously during part of the life cycle of each, combines spatial and temporal niche complementarities, and thus maximizes resource use. Roots of intercropped maize can enlarge the exploited soil volume to the rooting zone of previously harvested wheat, faba bean, chickpea or turnip and in this way benefit from nutrients and water remaining in the soil (Li *et al.*, 2011; Xia *et al.*, 2013b).

Spatial and temporal root complementarities have been attributed to the plasticity of root systems, i.e. ‘root decisions’ (Hodge, 2009), which adjust root location depending on nutrient availability and/or the presence of neighboring plants within a mono-species (Gersani *et al.*, 2001; Cahill *et al.*, 2010) or in intercropping (Hauggaard-Nielsen and Jensen, 2005; de Kroon, 2007; Cardinael *et al.*, 2015). How plants ‘make decisions’ to place their roots optimally for P acquisition in response to heterogeneity of the nutrient and the presence of neighboring plants is still a lively research topic (Croft *et al.*, 2012).

Biochemical niche complementarity can occur when two intercropped species tap into different pools of soil P (Fig. 1) such as inorganic and organic pools (Li *et al.*, 2008) or different chemical forms of each of these pools (Turner, 2008; Wang *et al.*, 2012). Wheat and associated white lupin plants in mixed culture were shown to deplete water-extractable and citric-acid-extractable inorganic P fractions in a low-P soil (4 parts washed sand, 1 part garden soil), respectively (Cu *et al.*, 2005). Chickpea was able to mobilize soil organic P and leave more inorganic P available to the intercropped wheat or maize in low-P sandy soils (Li *et al.*, 2003a; S. Li *et al.*, 2004). Analytical studies on P mobilization from different P pools in soil need to separate the rhizosphere of each intercropped species from the bulk soil to allow for accurate P fractionation and measurement of changes in the amounts of the various pools (Makoi *et al.*, 2010; Hinsinger *et al.*, 2011; Betencourt *et al.*, 2012; Tang *et al.*, 2014; Dissanayaka *et al.*, 2015).

Interspecific facilitation

Interspecific facilitation in P uptake occurs when one species increases soil P availability and the intercropped companion species can take advantage of that. Both direct root-mediated and indirect microbe-mediated processes are known to be involved in altering P availability in the rhizosphere under intercropping conditions (Fig. 1).

Direct root-mediated facilitation. Plant roots can alter P availability in the rhizosphere through P uptake or exudation of P-mobilizing/immobilizing compounds, or uptake of other nutrients such as calcium (Ca) (Hinsinger, 2001; Vance *et al.*, 2003; Devau *et al.*, 2010). Phosphorus mobilization in intercropping has been recorded for the insoluble inorganic and organic soil P pools through release of protons/hydroxyls, carboxylates or enzymes such as phosphatases and phytase (Fig. 1).

Intercropped cereals benefit from legumes in terms of P acquisition mostly in alkaline and neutral soils in which rhizosphere acidification in response to N₂ fixation raises P availability (Jensen, 1996; Hinsinger, 2001; Hauggaard-Nielsen *et al.*, 2009). Increased N₂ fixation in legumes as a consequence of competition of cereals for nitrate further stimulates P acquisition (Hinsinger *et al.*, 2011). Several experiments have shown a lower rhizosphere pH for legumes than cereals, while pH values in the rhizospheres of cereal-legume mixtures have been found to be intermediate (Li *et al.*, 2003a; Cu *et al.*, 2005; H. Li *et al.*, 2008). In addition, not only proton excretion and hence rhizosphere acidification but also reduced N₂ fixation and release of hydroxyl ions and consequent increase in rhizosphere pH can raise P availability in the rhizosphere of durum wheat and chickpea in a P-limited neutral soil (Betencourt *et al.*, 2012). This may occur as a consequence of changes in surface charges of (hydr)oxides and clay minerals to which phosphate ions are bound, and enhanced desorption of phosphate ions (Hinsinger, 2001). However, causal relationships between pH changes and changes in P availability and P uptake or plant growth in intercropping conditions remain to be established (Li *et al.*, 2008; Betencourt *et al.*, 2012).

Carboxylates including malate and citrate mobilize P from calcium phosphates in calcareous soils and P bound to oxides and hydroxides of aluminum (Al) and Fe in acid soils (Hinsinger, 2001). In calcareous soils, growing white lupin has been shown to increase P uptake of intercropped wheat because the roots of white lupin exude citrate which competes with phosphate ions for calcium phosphates, thereby increasing P dissolution and hence P availability (Gardner and Boundy, 1983). Roots of faba bean secrete protons, malate and citrate into the rhizosphere, which mobilize sparingly soluble P and contribute to increased P uptake by maize in P-deficient calcareous soils (Li *et al.*, 2007). Recent evidence indicates that malate exuded by intercropped faba bean diffuses to the roots of intercropped maize (H. Li *et al.*, 2014). The quantitative effects of carboxylate release on P solubility in the rhizosphere and P uptake by plants are of major current interest (Gerke, 2015); therefore, additional work is needed to ascertain the extent of chelation-based or carboxylate-driven P mobilization in intercropping systems.

Chickpea, a species that effectively accesses organic P from phytate by enzymatic hydrolysis, was shown to facilitate P acquisition of wheat (Li *et al.*, 2003a) and maize (S. Li *et al.*, 2004). No such effect was found when the source of supplied P was FePO₄ (Li *et al.*, 2003a). In contrast, Wang *et al.* (2007) observed no intercropping effect using another soil and different species and growth duration when P was added as phytate, whereas biomass, P acquisition, phosphatase activity and P availability all increased when FePO₄ was supplied. Thus, the differences in the intercropping effect may be related to crop cultivar, growth duration or soil type, all of which may determine the outcome of interactions between intercropped plants.

Calcium uptake by roots may release phosphate from calcium phosphates and thereby increase

soil P availability to stimulate plant P uptake (Devau *et al.*, 2010). Given that Ca uptake can differ substantially between species with legumes generally taking up more Ca than cereals, Hinsinger *et al.* (2011) speculated that more available P could be released from the rhizosphere of legumes than cereals to facilitate P acquisition of intercropped cereals. In addition, it is primarily the uptake of Ca cations that through compensatory release of protons leads to the soil acidification effect of rhizobial N₂ fixation in legume roots. Such acidification may also increase P availability in calcareous soils to facilitate P uptake by cereals. However, no detailed studies on this subject have been conducted in intercropping systems, which leaves this hypothesis to be validated.

Most cereal/legume intercropping studies implicitly assume that cereals shall benefit from the legumes because the roots of legumes are known to secrete larger amounts of protons (Tang *et al.*, 1997; Hinsinger *et al.*, 2003), carboxylates (Neumann and Römheld, 1999; Pearse *et al.*, 2006), and phosphatases (Nuruzzaman *et al.*, 2006; Makoi *et al.*, 2010) per unit of rhizosphere soil weight than those of cereals. However, cereals can also change rhizosphere P availability, and one might well expect the stimulation of P uptake in legumes in intercropping with P-efficient cereals or even two-way facilitation among cereals and legumes (Hinsinger *et al.*, 2011). Betencourt *et al.* (2012) recently reported that durum wheat also enhanced the P uptake and growth of intercropped chickpea through rhizosphere alkalization in a neutral soil. This occurs as a consequence of enhanced desorption of phosphate ions on charged surfaces of iron oxides and clay minerals (Hinsinger, 2001).

Indirect microbe-mediated facilitation. In addition to direct root-induced chemical processes, indirect interspecific P facilitation in the rhizosphere can occur as a consequence of shifts in microbial community structure, biomass or activity (He *et al.*, 2013; Tang *et al.*, 2014; Fig. 1). However, to our knowledge, there have been very few studies on P-solubilizing/mobilizing microorganisms (Richardson *et al.*, 2009; L. Zhang *et al.*, 2014), phytate-mobilizing bacteria (Jorquera *et al.*, 2008) and other organic-P-mineralizing microbes in intercropping conditions (He *et al.*, 2013). Theoretically, rhizodeposits may release P via stimulation of mineralization of soil organic matter (carbon priming) and contribute to increased P uptake in the companion crop. However, this mechanism has never been explored in cereal/legume intercropping (Hinsinger *et al.*, 2011), in which only the status of soil carbon and N stocks has been recently studied (Cong *et al.*, 2014, 2015). Direct transfer of nutrients between intercropped species via common mycorrhizal networks is better documented for N (Johansen and Jensen, 1996; He *et al.*, 2009) than for P (Li *et al.*, 2009; Merrild *et al.*, 2013; Qiao *et al.*, 2015).

Mechanistic modeling

Using an elegant modeling approach, Raynaud *et al.* (2008) demonstrated that only a few neighboring roots of non-exuding species can benefit from increased P availability as a consequence of citrate release from the species with root exudates because of its spatially limited diffusion in soil. Functional-structural simulation software was used to determine the effects of root architecture on nutrient uptake and biomass production of maize, bean and squash in monoculture, and maize/bean and maize/bean/squash mixtures in low-N and low-P soils during the first 40 days of root growth (Postma and Lynch, 2012). Postma and Lynch (2012) concluded

that spatial niche differentiation due to differences in root architecture enables over-yielding in mixtures, but direct competition for immobile P and direct facilitation in P uptake by maize in response to root exudates of bean or squash might be negligible because interspecies root spacing was found to be too large. These simulation studies provide testable predictions on spatial niche complementarity and interspecific facilitation of P uptake.

RHIZOSPHERE EFFECTS ON IRON AND ZINC UPTAKE BY LEGUME PLANTS INTERCROPPED WITH CEREALS

Observations

It has been demonstrated that intercropping legumes with cereals improved the micronutrient nutrition of legumes grown in both rhizobox and pot experiments and under field conditions. For example, intercropping with barley, oat, wheat or maize increased the chlorophyll, HCl-extractable Fe (so-called ‘active Fe’) and total Fe concentrations in young leaves of peanut, thus alleviating Fe chlorosis in calcareous soils (Zuo *et al.*, 2000, 2003; Inal *et al.*, 2007; Zuo and Zhang, 2008, 2009; Fig. 2A–B). It is interesting that the active Fe and chlorophyll concentrations in young leaves of peanut were closely (negatively) related with the distance to neighboring maize plants in the field (Zuo *et al.*, 2000). In addition, Fe and also Zn concentrations in the whole shoot of peanut were raised by co-culture of peanut with barley, oat, wheat or maize in calcareous sandy soils in a glasshouse (Zuo and Zhang, 2008). Finally, the improvement of Fe and Zn nutrition led to an increase in Fe and Zn concentrations in grains of legumes in intercropping systems of maize/peanut grown in a calcareous sandy soil (Zuo *et al.*, 2000; Zuo and Zhang, 2009) and wheat/chickpea in a calcareous soil (Gunes *et al.*, 2007).

Molecular and physiological processes in the rhizosphere in intercropping

Plant species have evolved different physiological mechanisms to increase Fe and Zn uptake and these are summarized by the terms ‘Strategy I’ and ‘Strategy II’ (see Marschner, 2011). The current biochemical model of Fe and Zn acquisition in intercropping (Fig. 3) is thus that strategy II supplements strategy I of Fe and Zn uptake in legume crops intercropped with cereals. For instance, when peanut (a Strategy I plant) and maize (a Strategy II plant) grow together in a calcareous sandy soil, phytosiderophores (PS, high-affinity Fe(III) chelators) secreted by maize roots help to solubilize Fe(III) in form of Fe(III)-PS complexes. Fe(III)-PS may diffuse from the root surface of maize to that of peanut. At the root apoplast of peanut, it may be then reduced to Fe(II) which is taken up by the Fe-deficient peanut. The evidence from molecular studies supports this reduction-based PS facilitation hypothesis in peanut/maize intercropping, in which the transcript levels of the gene encoding a Fe(III)-chelate reductase (*AhFRO1*) and genes encoding Fe(II) transporters (*AhIRT1*, *AhNRAMP1* and *AhDMT1*) were always higher in the roots of intercropped peanut than in the roots of monocropped peanut (Ding *et al.*, 2009, 2010; Xiong *et al.*, 2012; Guo *et al.*, 2014; Shen *et al.*, 2014; Fig. 3). However, there is still lack of direct evidence that intercropped peanut reduced PS-chelated Fe(III) to Fe(II) in the apoplast for Fe uptake. Deoxymugineic acid (DMA) is a type of PS secreted by maize roots and was detected in the rhizosphere of peanut plants intercropped with maize, and a Fe(III)-DMA transporter was expressed on the peanut root surface (Xiong *et al.*, 2013a; Figs. 2C and 3). This suggests that interspecific facilitation of Fe uptake is also likely to result from direct uptake of Fe(III)-DMA

by the intercropped peanut. However, the relative roles of these two (reduction-based and direct) uptake pathways of Fe(III)-PS are unclear.

Radio-isotopic experiments further support the PS-mediated facilitation of Fe uptake by intercropping of Strategy I and II plants. Cesco *et al.* (2006) found that Fe-deficient citrus plants increased ^{59}Fe -uptake from a dialysis tube to which suspension containing amorphous ^{59}Fe hydroxide was added in the presence of Fe-deficient barley plants or after addition of barley root exudates to the nutrient solution in hydroponics. Similarly, enhancement of ^{59}Fe uptake and alleviation of Fe chlorosis were also observed in Fe-deficient citrumelo 'Swingle' in the presence of the grasses *Poa pratensis* L. and *Festuca rubra* L. in solution culture (Cesco *et al.*, 2006; Fig. 2D-i). Leaf re-greening is an indication of Fe uptake and was not observed when citrumelo 'Swingle' and the *yellow stripe 3* (*ys3*) maize mutant (not able to release PS) were co-cultivated (Cesco *et al.*, 2006; Fig. 2D-ii).

Phytosiderophores are highly specific for Fe, but if present in excess they may also form quite strong complexes with Zn and other micronutrients. Transgenic plants and mutants over-expressing Fe(III) reductases and Fe(II) transporters also show an increased Zn accumulation (Zhu *et al.*, 2007). For example, the expression of barley *HvNAS1* in tobacco (*Nicotiana tabacum*) doubled the Fe and Zn concentrations in leaves (Takahashi *et al.*, 2003). It is therefore speculated that PS-mediated direct interspecific facilitation of Zn also occurs in intercropping of Strategy I and II plants (Fig. 3).

The experimental results about the effect of intercropping on PS release are inconsistent. Ding *et al.* (2009) and Xiong *et al.* (2013a) recorded substantially larger amounts of PS secreted by roots of maize intercropped with peanut than that of monocropped maize grown in calcareous sandy soils. In contrast, Inal *et al.* (2007) found the PS concentrations in the rhizosphere solution samples of maize were decreased by intercropping with peanut in a calcareous soil. The experimental results regarding the transcript levels of genes in relation to the reduction of Fe(III)-PS to Fe(II) for uptake by intercropped peanut were also inconsistent. Ding *et al.* (2009, 2010) showed that when Fe deficiency symptoms appeared in mono-cropped peanut, transcript levels of the Fe(III)-chelate reductase gene *AhFRO1* and Fe^{2+} -regulated transporter gene *AhIRT1* were higher in roots of intercropped than mono-cropped peanut in glasshouse pot experiments. However, Xiong *et al.* (2013b) found the expression of Fe-acquisition genes (mRNA) including both *AhFRO1* and *AhIRT1* were significantly upregulated by monocropping in glasshouse pot experiments. These conflicting findings may be explained by differences in the growth stages and conditions or Fe supplies (Table 2). For instance, Guo *et al.* (2014) found that the expression levels of *AhFRO1* and *AhYSL1* (the yellow stripe1-like family transporting Fe(III)-DMA gene) were higher in intercropping from day 46 to day 53, while their expressions were significantly lower than those in monocropping at day 69 when the plants were grown in pot experiments under greenhouse conditions.

Root and microbial responses to intercropping

Our understanding at the molecular and physiological levels has recently undergone major advances (Briat *et al.*, 2015). However, the roles of root morphology and microbes of the rhizosphere in Fe and Zn acquisition in intercropping remain poorly studied and understood (Table 2). Zuo *et al.* (2003) highlighted the possibility that root morphological changes and shifts

in composition and structure of microbial communities may also play a critical role in intercropping-facilitated Fe uptake. Pot experiments have demonstrated that co-culturing peanut with maize in Fe-deficient soils often increases the length and the number of lateral roots and root hairs of peanut. Simultaneously, rhizodermal transfer cells are formed in the subapical zone of peanut roots in response to intercropping (Zuo *et al.*, 2003). Intercropping decreased microbial abundance and activity in the rhizosphere of peanut grown in a calcareous sandy soil; Zuo *et al.* (2003) speculated that this may slow the degradation of mucous layers containing considerable amounts of root exudates, including PS. The thickness of mucous layers on the root surface of peanut was indeed increased by intercropping (Zuo *et al.*, 2003). These root morphological and microbial responses to intercropping may increase Fe uptake by intercropped peanut through increases in root surface area for Fe absorption and in PS-mediated interspecific facilitation.

Cluster roots of *Banksia attenuata* L. facilitate the acquisition of P and manganese (Mn) by the neighboring shrub (*Scholtzia involucrata* L. colonized by arbuscular mycorrhizal (AM) fungi) by making the nutrients more available for their neighbors in soils which consist of oligo-trophic coarse sand of low water-holding capacity and are severely nutrient impoverished and strongly leached (Muler *et al.*, 2014). The dual AM/ectomycorrhizal (EM) host *Melaleuca preissiana* L. exhibited enhanced EM colonization, favored EM over AM fungi and increased concentrations of P, Mn, Ca, Fe and boron (B) in the shoot when interacting with both the EM host *Eucalyptus marginata* L. and a non-mycorrhizal nutrient-mining plant with cluster roots (*Banksia menziesii* L.) in microcosms in which mesh barriers prevented the intermingling of roots (Teste *et al.*, 2014). The results show that a synergistic effect between EM hyphal scavenging and mobilization of limiting nutrients by cluster roots played a role in the observed nutrient facilitation. One may also speculate that facilitative uptake of micronutrients by legumes intercropped with cereals occurs via the hyphae of mycorrhizal fungi or rhizobia, accessing the nutrients mobilized by the roots of companion plants and/or by microbes (Jin *et al.*, 2014). Bacteria also release siderophores and other compounds that mobilize Fe from minerals. Interactions among crop plants, bacteria, and soil organic substances were shown to promote the formation of soluble Fe³⁺-complexes and thereby Fe availability for crop plants (Colombo *et al.*, 2014). Studies on the effects of microbes in the rhizosphere on Fe and Zn nutrition in intercropping are virtually non-existent. We know of only one report showing that there was no significant effect of soil sterilization on the Fe nutrition of peanut intercropped with maize in calcareous sandy soils (Fang *et al.*, 2004).

More to be learned from laboratory to field

Most studies demonstrating improved Fe and Zn nutrition of legume crops in response to the presence of cereals have been carried out in pots, rhizoboxes or hydroponic culture in the glasshouse (Fig. 2; Table 2). Studies in the field can yield quite different results from glasshouse experiments (Figs. 2 and 4; Table 2). In fact, intercropping was found to be unable to correct lime-induced Fe chlorosis in peanut on a poorly-aerated calcareous soil (Fig. 4A). In pot experiments, maize barely grew beyond the seedling or elongation stage and intercropping maize only slightly affected the growth of companion peanut in calcareous soils. Root, shoot and grain weights of short-lived peanut were thus rarely decreased substantially by co-cultivated maize and over-yielding even occurred (Table 2). However, these effects were not observed in the field

where the tall maize plants shaded the short-stemmed peanut or soybean to decrease biomass production of the latter (Fig. 4B–G). Shading was found to impair nutrient acquisition in intercropping systems of wheat (*Triticum turgidum durum*) with alfalfa (*Medicago sativa*) or soybean (Whitehead and Isaac, 2012), and of maize with soybean (Gong *et al.*, 2015), mainly due to reduced photosynthetic capacity and biomass accumulation of the shaded crop as compared to monocropping.

RELATIONSHIPS BETWEEN INTERSPECIFIC ROOT INTERACTIONS AND ABOVE-GROUND BIOMASS AND NUTRIENT ACCUMULATION

Mass fluxes of water, nutrients and photosynthetic organic materials between above-ground and below-ground components of crops are influenced by intercropping (Malézieux *et al.*, 2009). It has been reported that below-ground (root) interactions played a more important role than above-ground (shoot) interactions in crop growth and intercropping advantages in maize/potato intercropping (Wu *et al.*, 2012). The present review suggests that both interspecific root physiology (e.g. root secretion of carboxylates and PS) and morphology (e.g. length and distribution of roots in soil profiles) contribute to plant acquisition of P, Fe and Zn from sparingly soluble sources in soils. It is difficult to quantify the actual contribution of root physiology to shoot biomass and to P, Fe and Zn accumulation, but research continues in an attempt to evaluate the relationships between the interspecific root biomass and its distribution and above-ground biomass and nutrient accumulation, especially in diverse plant communities.

Root over-production in mixtures has been suggested to contribute to increased total biomass production in diverse plant communities over time through effects on the availability and acquisition of nutrients in soil (Tilman *et al.*, 1996; de Kroon *et al.*, 2012). The total productivity of diverse plant communities may also partly depend on below-ground interactions that cause overall roots to be distributed deeply in soil profiles. Mueller *et al.* (2013) evaluated the links among diversity, overall root depth distribution and total productivity using data from a 12-year experiment, and observed that the overall proportion of deep-root biomass increased with species richness and was positively correlated with above-ground productivity in artificial grasslands. Mommer *et al.* (2015) recently showed that diversity enhanced overall root length and decreased total root loss and root mortality of the whole community, resulting in root over-production in an experimental grassland community. These results allow us to reaffirm that the total productivity and the use of resources by diverse plant communities depend on overall production of roots or root distribution in the soil profile.

Indeed, the contributions of individual species in diverse plant communities are not just additive in comparison with monoculture systems, but also result in positive or negative interaction effects on root biomass production, root distribution, nutrient acquisition and above-ground productivity, which need further investigation. Across eleven monocropped grassland plant species (i.e. legumes, high-nitrogen rhizomatous C₃ species and a separate gradient of differentiation from tall- to short-stature species) and soil depth (0-95 cm) examined, strong positive correlations were observed between the density of fine roots and depletion of soil water and N, but there were no relationships with the biomass of coarse roots (Craine *et al.*, 2003). Thus, we speculate that the differentiation of coarse and fine root biomass between monoculture and diverse plant communities also plays an important role in the relationship

between plant diversity and nutrient acquisition/biomass production.

Compared to the above-mentioned diverse plant communities (e.g. grassland systems), very few studies have been conducted to evaluate relationships between interspecific root interactions and nutrient acquisition/biomass productivity as affected by intercropping of cereals with legume crops, especially under field conditions (Dordas *et al.*, 2012; Wu *et al.*, 2012). It appears that there are positive links between root growth space and root length density of the individual crop species and the corresponding above-ground growth in intercropping systems, but there is a lack of direct evidence and statistically significant data (Li *et al.*, 2001a, b, 2006, 2011). A recent study reported ‘competition–recovery–over-yielding’ dynamics in root length density, root growth space and corresponding shoot biomass of maize intercropped with faba bean or chickpea in a calcareous soil under field conditions (Xia *et al.*, 2013b). The total root length and root growth space of intercropped maize on a per-plant basis was initially lower than (at the maize elongation stage) and then equal to (at the maize tasseling stage) and finally larger than (after earlier maturing legume crop harvest at maize grain-filling stage) in monocropped maize. The same was also found in maize shoot growth on a per-plant basis, and the dynamics of root length and root growth space correlated well with the dynamics of shoot biomass of maize (Xia *et al.*, 2013b). The relationships between responses of root system and shoot growth and the uptake of P, Fe and Zn in cereal/legume intercropping need further elucidation.

INTERCROPPING AFFECTS NUTRIENT ALLOCATION IN CROP PLANTS

Intercropping influences nutrient distribution among different plant parts or translocation of nutrients from root to shoot and/or from shoot to grains, but the effects vary, depending on types of nutrient elements and crop species. For example, in a pot experiment Xiao *et al.* (2013) found that Fe concentrations decreased in the shoots but increased in the roots of cucumber in intercropping with green garlic compared to monocropping in an alkaline soil, indicating that intercropping decreased the translocation of Fe from roots to shoots. In a field experiment with wheat/chickpea intercropping, intercropping stimulated Fe translocation from shoots to seeds of chickpea, showing higher Fe concentrations in seeds but not in shoots than monocropping. Simultaneously, intercropping decreased the translocation of Zn from shoots to seeds of wheat, showing higher Zn concentrations in the shoot but not in grains (Gunes *et al.*, 2007).

Our latest findings show that although intercropping stimulated total uptake of Fe, Mn, copper (Cu) and Zn in the above-ground parts of maize, it decreased the concentrations of these nutrients in the grains due to reduced translocation of the nutrients from the vegetative tissues to the grains (Xia *et al.*, 2013c). Therefore, grain nutritional status in intercropping systems is not only determined by facilitation effects on the overall uptake of individual nutrients from soil but also by the translocation of nutrients to grains via the xylem and phloem within plants.

According to experimental results from single-species cropping of wheat, a dilution effect was observed in relation to the concentrations of Zn, Fe, Cu and Mn in grains, which were negatively correlated with grain yields (Jarrell and Beverly, 1981; Fan *et al.*, 2008; Zhao and McGrath, 2009). Higher grain yields of cereals on a per-plant basis in intercropping than in monocropping may also lead to a dilution effect. For instance, in a maize-based intercropping experiment with turnip, faba bean, chickpea and soybean at various P application rates, grain yields of intercropped maize were significantly higher than those of monocropped maize on a per-plant

basis, but the concentrations (on a dry-weight basis) of Fe and Cu in grains were negatively correlated with grain yields across the treatments (Xia *et al.*, 2013c).

Intercropping may also affect the interactions (synergism and antagonism) between mineral elements, which in turn affect nutrient allocation between plant parts. For example, there is a strong antagonism between P and Zn which affects their uptake from soil, and their mobility within plants (Warnock, 1970; Zhu *et al.*, 2002). In a study by Inal *et al.* (2007), intercropping increased the concentrations of Fe, Zn, P and potassium (K) in shoots of intercropped peanut and maize but lowered the concentration of Ca, and did not affect those of N and Mn. Xiao *et al.* (2013) found that the concentrations of N, P, K, Ca and Mn in cucumber shoots and roots were significantly higher but the magnesium (Mg) concentrations were lower in intercropping with green garlic than monocropping. The quantitative relationships among nutrients in intercropping appear to be different from those in monocropping. Further study is needed in this respect.

CONCLUSIONS AND OUTLOOK

Phosphorus, Fe and Zn nutrition of cereals and/or legumes can be improved by intercropping the two crops through the increased availability of P, Fe and Zn in the rhizosphere (Fig. 5). Thorough soil exploitation by complementarity of root systems in space and time in intercropping allows for an efficient use of P, Fe and Zn in soil. The increased release of exudates (e.g. protons, carboxylates, phosphatases and PS) from crop plant roots and soil microbes under intercropping may increase the solubility of P, Fe and Zn which can benefit both intercropped cereals and legumes. Adaptive root-microbe responses in intercropping enable flexible improvements of crops to exploit sparingly soluble nutrients in soil. Much has been learned at the molecular and whole soil-plant levels about P, Fe and Zn acquisition in single-species cropping. However, our understanding with respect to the foraging for these nutrients by roots and microbes and nutrient allocation to different plant tissues is rather limited for intercropping systems (Fig. 5).

This review has focused on P acquisition advantages in the whole cereal/legume intercropping system (Fig. 1) and on PS-mediated interspecific facilitation of Fe and Zn uptake by legumes intercropped with cereals (Fig. 3). By bringing these two aspects together, we identify the value of studies on P and on Fe/Zn uptake in intercropping in informing each other. Interspecific facilitation of Fe/Zn uptake occurring in the direction of cereals to legumes has been supported by evidence from studies using radio-isotopic tracing (Cesco *et al.*, 2006) and molecular biotechnology (Xiong *et al.*, 2013a). However, there is no direct mechanistic evidence for interspecific facilitation of P uptake. Isotopic and molecular physiological methods need to be applied to understand the nature of interspecific P facilitation. Niche complementarity may also contribute to increasing Fe and Zn acquisition in cereal-legume intercropping systems, an aspect not fully understood that needs to be explored in future research. The relative importance of niche complementarity and interspecific facilitation for nutrient acquisition should be quantified for various intercropping systems so that complementarity and facilitation can be manipulated to increase P and Fe/Zn acquisition in low- and high-input agricultural ecosystems.

Future research is also warranted in the following aspects. (1) Interactions between root growth and microbes on plant acquisition of P, Fe and Zn under intercropping need to be further explored (Tables 1 and 2). Such studies should be expanded to combinations of new crop species

and soil types, particularly acid soils and in field situations. (2) Time-course studies in intercropping systems will further elucidate the dynamics of P, Fe and Zn mobilization and acquisition from soil, and their association with translocation of the nutrients from roots to shoots and to grains. (3) Use of multi-disciplinary approaches, covering soil science, plant physiology, agronomy and ecology, may help to design effective intercropping systems that maximize the use of P, Fe and Zn from soil available pools and accumulation of Fe and Zn in grains together with appropriate farming practices. In this respect, all components including crop species, farming practices and environmental conditions must be considered together in a holistic fashion as shown in Fig. 5.

ACKNOWLEDGEMENTS

We thank anonymous reviewers and the handling editor for their constructive comments on the paper. Funding was provided by the National Natural Science Foundation of China (31501834), the High-Level Talents and Innovative Team Recruitment Program of the Shandong Academy of Agricultural Sciences, the Young Scientist Research Foundation of the Shandong Academy of Agricultural Sciences (2014QNM07), the Innovation Projects for Key Agricultural Technology Application for Shandong Province in China: High Efficient Use of Fertilizer and Water in the Rotation System of Wheat-Maize Intercropping with Peanut (2014), and by the Shandong Modern Agricultural Technology and Industry System.

LITERATURE CITED

- Amossé C, Jeuffroy MH, Mary B, David C. 2014.** Contribution of relay intercropping with legume cover crops on nitrogen dynamics in organic grain systems. *Nutrient Cycling in Agroecosystems* **98**: 1–14.
- Bainard LD, Koch AM, Gordon AM, Klironomos JN. 2012.** Temporal and compositional differences of arbuscular mycorrhizal fungal communities in conventional monocropping and tree-based intercropping systems. *Soil Biology and Biochemistry* **45**: 172–180.
- Betencourt E, Duputel M, Colomb B, Desclaux D, Hinsinger P. 2012.** Intercropping promotes the ability of durum wheat and chickpea to increase rhizosphere phosphorus availability in a low P soil. *Soil Biology and Biochemistry* **46**: 181–190.
- Bevis LEM. 2015.** Soil-to-human mineral transmission with an emphasis on zinc, selenium, and iodine. *Springer Science Reviews* **3**: 77–96.
- Briat JF, Dubos C, Gaymard F. 2015.** Iron nutrition, biomass production, and plant product quality. *Trends in Plant Science* **20**: 33–40.
- Brooker RW, Bennett AE, Cong WF, et al. 2015.** Improving intercropping: a synthesis of research in agronomy, plant physiology and ecology. *New Phytologist* **206**: 107–117.
- Cahill JF, McNickle GG, Haag JJ, Lamb EG, Nyanumba SM, Clair CCS. 2010.** Plants integrate information about nutrients and neighbors. *Science* **328**: 1657–1657.
- Callaway RM. 1995.** Positive interactions among plants. *The Botanical Review* **61**: 306–349.
- Cardinael R, Mao Z, Prieto I, et al. 2015.** Competition with winter crops induces deeper rooting of walnut trees in a Mediterranean alley cropping agroforestry system. *Plant and Soil*, in press. <http://dx.doi.org/10.1007/s11104-015-2422-8>.
- Cesco S, Rombola AD, Tagliavini M, Varanini Z, Pinton R. 2006.** Phytosiderophores released by graminaceous species promote ⁵⁹Fe-uptake in citrus. *Plant and Soil* **287**: 223–233.
- Colombo C, Palumbo G, He JZ, Pinton R, Cesco S. 2014.** Review on iron availability in soil: interaction of Fe

- minerals, plants, and microbes. *Journal of Soils and Sediments* **14**: 538–548.
- Cong WF, Hoffland E, Li L, Janssen BH, van der Werf W. 2015. Intercropping affects the rate of decomposition of soil organic matter and root litter. *Plant and Soil* **391**: 399–411.
- Cong WF, van Ruijven J, Mommer L, De Deyn GB, Berendse F, Hoffland E. 2014. Plant species richness promotes soil carbon and nitrogen stocks in grasslands without legumes. *Journal of Ecology* **102**: 1163–1170.
- Cornu JY, Deinlein U, HÖreth S, *et al.* 2015. Contrasting effects of nicotianamine synthase knockdown on zinc and nickel tolerance and accumulation in the zinc/cadmium hyperaccumulator *Arabidopsis halleri*. *New Phytologist* **206**: 738–750.
- Craine J, Wedin D, Chapin F, Reich P. 2003. Relationship between the structure of root systems and resource use for 11 North American grassland plants. *Plant Ecology* **165**: 85–100.
- Croft S, Hodge A, Pitchford J. 2012. Optimal root proliferation strategies: the roles of nutrient heterogeneity, competition and mycorrhizal networks. *Plant and Soil* **351**: 191–206.
- Cu ST, Hutson J, Schuller KA. 2005. Mixed culture of wheat (*Triticum aestivum* L.) with white lupin (*Lupinus albus* L.) improves the growth and phosphorus nutrition of the wheat. *Plant and Soil* **272**: 143–151.
- de Kroon H. 2007. How do roots interact? *Science* **318**: 1562–1563.
- de Kroon H, Hendriks M, van Ruijven J, *et al.* 2012. Root responses to nutrients and soil biota: drivers of species coexistence and ecosystem productivity. *Journal of Ecology* **100**: 6–15.
- Dessougi HE, Dreele AZ, Claassen N. 2003. Growth and phosphorus uptake of maize cultivated alone, in mixed culture with other crops or after incorporation of their residues. *Journal of Plant Nutrition and Soil Science* **166**: 254–261.
- Devau N, Le Cadre E, Hinsinger P, Gérard F. 2010. A mechanistic model for understanding root-induced chemical changes controlling phosphorus availability. *Annals of Botany* **105**: 1183–1197.
- Dey AK. 2003. Effect of phosphorus fertilization on quality parameters of soybean+maize intercropping system in the Tarai soils of Uttaranchal. *Crop Research-Hisar* **26**: 374–377.
- Ding H, Duan L, Wu H, *et al.* 2009. Regulation of AhFRO1, an Fe(III)-chelate reductase of peanut, during iron deficiency stress and intercropping with maize. *Physiologia Plantarum* **136**: 274–283.
- Ding H, Duan L, Li J, *et al.* 2010. Cloning and functional analysis of the peanut iron transporter *AhIRT1* during iron deficiency stress and intercropping with maize. *Journal of Plant Physiology* **167**: 996–1002.
- Dissanayaka DMSB, Maruyama H, Masuda G, Wasaki J. 2015. Interspecific facilitation of P acquisition in intercropping of maize with white lupin in two contrasting soils as influenced by different rates and forms of P supply. *Plant and Soil* **390**: 223–236.
- Dordas CA, Vlachostergios DN, Lithourgidis AS. 2012. Growth dynamics and agronomic-economic benefits of pea-oat and pea-barley intercrops. *Crop and Pasture Science* **63**: 45–52.
- Dragicevic V, Oljaca S, Stojiljkovic M, Simic M, Dolijanovic Z, Kravic N. 2015. Effect of the maize-soybean intercropping system on the potential bioavailability of magnesium, iron and zinc. *Crop and Pasture Science*, in press.
- Ehrmann J, Ritz K. 2013. Plant: soil interactions in temperate multi-cropping production systems. *Plant and Soil* **376**: 1–29.
- Fan MS, Zhao FJ, Fairweather-Tait SJ, Poulton PR, Dunham SJ, McGrath SP. 2008. Evidence of decreasing mineral density in wheat grain over the last 160 years. *Journal of Trace Elements in Medicine and Biology* **22**: 315–324.
- Fang ZG, Zuo YM, Li L, FS Zhang. 2004. Effects of mixed cropping of maize and peanut on the Fe nutrition of peanut in sterilized soil. *Chinese Journal of Eco-Agriculture* **12**: 98–101. [in Chinese]

- Fridley JD. 2001.** The influence of species diversity on ecosystem productivity: how, where, and why? *Oikos* **93**: 514–526.
- Gardner WK, Boundy KA. 1983.** The acquisition of phosphorus by *Lupinus albus* L. IV. The effect of interplanting wheat and white lupin on the growth and mineral composition of the two species. *Plant and Soil* **70**: 391–402.
- 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.
- Gersani M, O'Brien EE, Maina GM, Abramsky Z. 2001.** Tragedy of the commons as a result of root competition. *Journal of Ecology* **89**: 660–669.
- Gong WZ, Jiang CD, Wu YS, Chen HH, Liu WY, Yang WY. 2015.** Tolerance vs. avoidance: two strategies of soybean (*Glycine max*) seedlings in response to shade in intercropping. *Phytosynthetica* **53**: 259–268.
- Gunes A, Inal A, Adak MS, et al. 2007.** Mineral nutrition of wheat, chickpea and lentil as affected by mixed cropping and soil moisture. *Nutrient Cycling in Agroecosystems* **78**: 83–96.
- Guo X, Xiong H, Shen H, et al. 2014.** Dynamics in the rhizosphere and iron-uptake gene expression in peanut induced by intercropping with maize: role in improving iron nutrition in peanut. *Plant Physiology and Biochemistry* **76**: 36–43.
- Hauggaard-Nielsen H, Jensen ES. 2005.** Facilitative root interactions in intercrops. *Plant and Soil* **274**: 237–250.
- Hauggaard-Nielsen H, Gooding M, Ambus P, et al. 2009.** Pea–barley intercropping for efficient symbiotic N₂-fixation, soil N acquisition and use of other nutrients in European organic cropping systems. *Field Crops Research* **113**: 64–71.
- He XH, Critchley C, Nara K, Southworth D, Bledsoe CS. 2009.** ¹⁵N enrichment methods to quantify two-way nitrogen transfer between plants linked by mycorrhizal networks. *Symbiotic Fungi Soil Biology* **18**: 285–291.
- He Y, Ding N, Shi J, Wu M, Liao H, Xu J. 2013.** Profiling of microbial PLFAs: implications for interspecific interactions due to intercropping which increase phosphorus uptake in phosphorus limited acidic soils. *Soil Biology and Biochemistry* **57**: 625–634.
- Hinsinger P. 2001.** Bioavailability of soil inorganic P in the rhizosphere as affected by root-induced chemical changes: a review. *Plant and Soil* **237**: 173–195.
- Hinsinger P, Plassard C, Tang C, Jaillard B. 2003.** Origins of root-mediated pH changes in the rhizosphere and their responses to environmental constraints: a review. *Plant and Soil* **248**: 43–59.
- Hinsinger P, Betencourt E, Bernard L, et al. 2011.** P for two, sharing a scarce resource: soil phosphorus acquisition in the rhizosphere of intercropped species. *Plant Physiology* **156**: 1078–1086.
- Hodge A. 2009.** Root decisions. *Plant, Cell and Environment* **32**: 628–640.
- Horst W, Waschkes C. 1987.** Phosphorus nutrition of spring wheat (*Triticum aestivum* L.) in mixed culture with white lupine (*Lupinus albus* L.). *Zeitschrift für Pflanzenernährung und Bodenkunde* **150**: 1–8.
- Inal A, Gunes A, Zhang F, Cakmak I. 2007.** Peanut/maize intercropping induced changes in rhizosphere and nutrient concentrations in shoots. *Plant Physiology and Biochemistry* **45**: 350–356.
- Jarrell WM, Beverly RB. 1981.** The dilution effect in plant nutrition studies. *Advances in Agronomy* **34**: 197–224.
- Jensen ES. 1996.** Grain yield, symbiotic N₂ fixation and interspecific competition for inorganic N in pea-barley intercrops. *Plant and Soil* **182**: 25–38.
- Jiao NY, Hou LT, Ning TY, Wang JT, Zhang PF, Fu GZ. 2012.** The effects of different genotype maize on Fe nutrition of intercropped peanut. *Journal of Peanut Science* **41**: 8–11. [in Chinese]

- Jin CW, Ye YQ, Zheng SJ. 2014.** An underground tale: contribution of microbial activity to plant iron acquisition via ecological processes. *Annals of Botany* **113**: 7–18.
- Johansen A, Jensen ES. 1996.** Transfer of N and P from intact or decomposing roots of pea to barley interconnected by an arbuscular mycorrhizal fungus. *Soil Biology and Biochemistry* **28**: 73–81.
- Jorquera MA, Hernández MT, Rengel Z, Marschner P, de la Luz Mora M. 2008.** Isolation of culturable phosphobacteria with both phytate-mineralization and phosphate-solubilization activity from the rhizosphere of plants grown in a volcanic soil. *Biology and Fertility of Soils* **44**: 1025–1034.
- Lelei JJ, Onwonga RN. 2014.** White lupin (*Lupinus albus* L. cv. Amiga) increases solubility of Minjingu phosphate rock, phosphorus balances and maize yields in Njoro Kenya. *Sustainable Agriculture Research* **3**: 37–49.
- Li H, Shen J, Zhang F, et al. 2008.** Dynamics of phosphorus fractions in the rhizosphere of common bean (*Phaseolus vulgaris* L.) and durum wheat (*Triticum turgidum durum* L.) grown in monocropping and intercropping systems. *Plant and Soil* **312**: 139–150.
- Li H, Zhang F, Rengel Z, Shen J. 2014.** Rhizosphere properties in monocropping and intercropping systems between faba bean (*Vicia faba* L.) and maize (*Zea mays* L.) grown in a calcareous soil. *Crop and Pasture Science* **64**: 976–984.
- Li L, Yang S, Li X, Zhang F, Christie P. 1999.** Interspecific complementary and competitive interactions between intercropped maize and faba bean. *Plant and Soil* **212**: 105–114.
- Li L, Sun J, Zhang F, Li X, Rengel Z, Yang S. 2001a.** Wheat/maize or wheat/soybean strip intercropping: II. Recovery or compensation of maize and soybean after wheat harvesting. *Field Crops Research* **71**: 173–181.
- Li L, Sun J, Zhang F, Li X, Yang S, Rengel Z. 2001b.** Wheat/maize or wheat/soybean strip intercropping: I. Yield advantage and interspecific interactions on nutrients. *Field Crops Research* **71**: 123–137.
- Li L, Tang C, Rengel Z, Zhang F. 2003a.** Chickpea facilitates phosphorus uptake by intercropped wheat from an organic phosphorus source. *Plant and Soil* **248**: 297–303.
- Li L, Zhang F, Li X, et al. 2003b.** Interspecific facilitation of nutrient uptake by intercropped maize and faba bean. *Nutrient Cycling in Agroecosystems* **65**: 61–71.
- Li L, Sun J, Zhang F, et al. 2006.** Root distribution and interactions between intercropped species. *Oecologia* **147**: 280–290.
- Li L, Li SM, Sun JH, et al. 2007.** Diversity enhances agricultural productivity via rhizosphere phosphorus facilitation on phosphorus-deficient soils. *Proceedings of the National Academy of Sciences, USA* **104**: 11192–11196.
- Li L, Sun J, Zhang F. 2011.** Intercropping with wheat leads to greater root weight density and larger below-ground space of irrigated maize at late growth stages. *Soil Science and Plant Nutrition* **57**: 61–67.
- Li L, Tilman D, Lambers H, Zhang FS. 2014.** Plant diversity and overyielding: insights from belowground facilitation of intercropping in agriculture. *New Phytologist* **203**: 63–69.
- Li S, Li L, Zhang F, Tang C. 2004.** Acid phosphatase role in chickpea/maize intercropping. *Annals of Botany* **94**: 297–303.
- Li Y, Ran W, Zhang R, Sun S, Xu G. 2009.** Facilitated legume nodulation, phosphate uptake and nitrogen transfer by arbuscular inoculation in an upland rice and mung bean intercropping system. *Plant and Soil* **315**: 285–296.
- Lithourgidis AS, Vlachostergios DN, Dordas CA, Damalas CA. 2011.** Dry matter yield, nitrogen content, and competition in pea-cereal intercropping systems. *European Journal of Agronomy* **34**: 287–294.
- Makoi JHJR, Chimphango SBM, Dakora FD. 2010.** Elevated levels of acid and alkaline phosphatase activity in roots and rhizosphere of cowpea (*Vigna unguiculata* L. Walp.) genotypes grown in mixed culture and at

- different densities with sorghum (*Sorghum bicolor* L.). *Crop and Pasture Science* **61**: 279–286.
- Malézieux E, Crozat Y, Dupraz C, et al. 2009.** Mixing plant species in cropping systems: concepts, tools and models: a review. *Agronomy for Sustainable Development* **29**: 43–62.
- Marschner P. 2011.** *Marschner's mineral nutrition of higher plants*, 3rd edn. London: Academic Press.
- Mei PP, Gui LG, Wang P, et al. 2012.** Maize/faba bean intercropping with rhizobia inoculation enhances productivity and recovery of fertilizer P in a reclaimed desert soil. *Field Crops Research* **130**: 19–27.
- Merrild MP, Ambus P, Rosendahl S, Jakobsen I. 2013.** Comm arbuscular mycorrhizal networks amplify competition for phosphorus between seedlings and established plants. *New Phytologist* **200**: 229–240.
- Mommer L, Padilla FM, van Ruijven J, et al. 2015.** Diversity effects on root length production and loss in an experimental grassland community. *Functional Ecology*, in press. <http://dx.doi.org/10.1111/1365-2435.12466>.
- Mueller KE, Tilman D, Fornara DA, Hobbie SE. 2013.** Root depth distribution and the diversity-productivity relationship in a long-term grassland experiment. *Ecology* **94**: 787–793.
- Muler AL, Oliveira R, Lambers H, Veneklaas EJ. 2014.** Does cluster-root activity benefit nutrient uptake and growth of co-existing species? *Oecologia* **174**: 23–31.
- Musa E, Elsheikh E, Mohamed Ahmed I, Babiker E. 2011.** Effect of intercropping, *Bradyrhizobium* inoculation and N, P fertilizers on yields, physical and chemical quality of cowpea seeds. *Frontiers of Agriculture in China* **5**: 543–551.
- Neumann G, Römheld V. 1999.** Root excretion of carboxylic acids and protons in phosphorus-deficient plants. *Plant and Soil* **211**: 121–130.
- Nuruzzaman M, Lambers H, Bolland MD, Veneklaas EJ. 2006.** Distribution of carboxylates and acid phosphatase and depletion of different phosphorus fractions in the rhizosphere of a cereal and three grain legumes. *Plant and Soil* **281**: 109–120.
- Pearse SJ, Veneklaas EJ, Cawthray GR, Bolland MD, Lambers H. 2006.** Carboxylate release of wheat, canola and 11 grain legume species as affected by phosphorus status. *Plant and Soil* **288**: 127–139.
- Postma JA, Lynch JP. 2012.** Complementarity in root architecture for nutrient uptake in ancient maize/bean and maize/bean/squash polycultures. *Annals of Botany* **110**: 521–534.
- Qiao X, Bei S, Li C, et al. 2015.** Enhancement of faba bean competitive ability by arbuscular mycorrhizal fungi is highly correlated with dynamic nutrient acquisition by competing wheat. *Scientific Reports* **5**: 8122.
- Raynaud X, Jaillard B, Leadley PW. 2008.** Plants may alter competition by modifying nutrient bioavailability in rhizosphere: a modeling approach. *The American Naturalist* **171**: 44–58.
- Richardson AE, Barea JM, McNeill AM, Prigent-Combaret C. 2009.** Acquisition of phosphorus and nitrogen in the rhizosphere and plant growth promotion by microorganisms. *Plant and Soil* **321**: 305–339.
- Rose TJ, Rengel Z, Ma Q, Bowden JW. 2007.** Differential accumulation patterns of phosphorus and potassium by canola cultivars compared to wheat. *Journal of Plant Nutrition and Soil Science* **170**: 404–411.
- Sekiya N, Araki H, Yano K. 2011.** Applying hydraulic lift in an agroecosystem: forage plants with shoots removed supply water to neighboring vegetable crops. *Plant and Soil* **341**: 39–50.
- Shen H, Xiong H, Guo X, et al. 2014.** AhDMT1, a Fe²⁺ transporter, is involved in improving iron nutrition and N₂ fixation in nodules of peanut intercropped with maize in calcareous soils. *Planta* **239**: 1065–1077.
- Shen J, Li C, Mi G, et al. 2013.** Maximizing root/rhizosphere efficiency to improve crop productivity and nutrient use efficiency in intensive agriculture of China. *Journal of Experimental Botany* **64**: 1181–1192.
- Soltani S, Khoshgoftarmanesh A, Afyuni M, Shrivani M, Schulin R. 2014.** The effect of preceding crop on wheat grain zinc concentration and its relationship to total amino acids and dissolved organic carbon in rhizosphere soil solution. *Biology and Fertility of Soils* **50**: 239–247.

- Stein A. 2010.** Global impacts of human mineral malnutrition. *Plant and Soil* **335**: 133–154.
- Sun Y, Zhang N, Wang ET, Yuan H, Yang J, Chen W. 2013.** Influence of intercropping and intercropping plus rhizobial inoculation on microbial activity and community composition in rhizosphere of alfalfa (*Medicago sativa* L.) and Siberian wildrye (*Elymus sibiricus* L.). *FEMS Microbiology Ecology* **70**: 218–226.
- Takahashi M, Terada Y, Nakai I, et al. 2003.** Role of nicotianamine in the intracellular delivery of metals and plant reproductive development. *Plant Cell* **15**: 1263–1280.
- Tang C, Barton L, McLay C. 1997.** A comparison of proton excretion of twelve pasture legumes grown in nutrient solution. *Australian Journal of Experimental Agriculture* **37**: 563–570.
- Tang X, Bernard L, Brauman A, et al. 2014.** Increase in microbial biomass and phosphorus availability in the rhizosphere of intercropped cereal and legumes under field conditions. *Soil Biology and Biochemistry* **75**: 86–93.
- Teste FP, Veneklaas EJ, Dixon KW, Lambers H. 2014.** Complementary plant nutrient-acquisition strategies promote growth of neighbour species. *Functional Ecology* **28**: 819–828.
- Tilman D, Wedin D, Knops J. 1996.** Productivity and sustainability influenced by biodiversity in grassland ecosystems. *Nature* **379**: 718–720.
- Tilman D, Reich PB, Knops J, Wedin D, Mielke T, Lehman C. 2001.** Diversity and productivity in a long-term grassland experiment. *Science* **294**: 843–845.
- Tong Y, Gabriel-Neumann E, Krumbein A, Ngwene B, George E, Schreiner M. 2015.** Interactive effects of arbuscular mycorrhizal fungi and intercropping with sesame (*Sesamum indicum*) on the glucosinolate profile in broccoli (*Brassica oleracea* var. Italica). *Environmental and Experimental Botany* **109**: 288–296.
- Turner BL. 2008.** Resource partitioning for soil phosphorus: a hypothesis. *Journal of Ecology* **96**: 698–702.
- Vance CP, Uhde-Stone C, Allan DL. 2003.** Phosphorus acquisition and use: critical adaptations by plants for securing a nonrenewable resource. *New Phytologist* **157**: 423–447.
- Varanini Z, Cesco S, Monte R, Tomasi N, Pinton R. 2008.** Plant nutrition between chemical and physiological limitations: is a sustainable approach possible? *Italian Journal of Agronomy* **3**: 129–141.
- Walder F, Niemann H, Natarajan M, Lehmann MF, Boller T, Wiemken A. 2012.** Mycorrhizal networks: common goods of plants shared under unequal terms of trade. *Plant Physiology* **159**: 789–797.
- Wang D, Marschner P, Solaiman Z, Rengel Z. 2007.** Growth, P uptake and rhizosphere properties of intercropped wheat and chickpea in soil amended with iron phosphate or phytate. *Soil Biology and Biochemistry* **39**: 249–256.
- Wang Y, Marschner P, Zhang F. 2012.** Phosphorus pools and other soil properties in the rhizosphere of wheat and legumes growing in three soils in monoculture or as a mixture of wheat and legume. *Plant and Soil* **354**: 283–298.
- Warnock R. 1970.** Micronutrient uptake and mobility within corn plants (*Zea mays* L.) in relation to phosphorus-induced zinc deficiency. *Soil Science Society of America Journal* **34**: 765–769.
- Whitehead M, Isaac ME. 2012.** Effects of shade on nitrogen and phosphorus acquisition in cereal-legume intercropping systems. *Agriculture* **2**: 12–24.
- Wu K, Fullen MA, An T, et al. 2012.** Above- and below-ground interspecific interaction in intercropped maize and potato: A field study using the 'target' technique. *Field Crops Research* **139**: 63–70.
- Xia HY, Wang ZG, Zhao JH, et al. 2013a.** Contribution of interspecific interactions and phosphorus application to sustainable and productive intercropping systems. *Field Crops Research* **154**: 53–64.
- Xia HY, Zhao JH, Sun JH, et al. 2013b.** Dynamics of root length and distribution and shoot biomass of maize as affected by intercropping with different companion crops and phosphorus application rates. *Field Crops Research* **150**: 52–62.

- Xia HY, Zhao JH, Sun JH, et al. 2013c.** Maize grain concentrations and above-ground shoot acquisition of micronutrients as affected by intercropping with turnip, faba bean, chickpea, and soybean. *Science China Life Sciences* **56**: 823–834.
- Xiao XM, Cheng ZH, Meng HW, Liu LH, Li HZ, Dong YX. 2013.** Inetercropping of green garlic (*Allium sativum* L.) induces nutrient concentration changes in the soil and plants in continuously cropped cucumber (*Cucumis sativus* L.) in a plastic tunnel. *PLoS One* **8**: e62173. <http://dx.doi.org/10.1371/journal.pone.0062173>.
- Xiong H, Kobayashi T, Kakei Y, et al. 2012.** AhNRAMP1 iron transporter is involved in iron acquisition in peanut. *Journal of Experimental Botany* **63**: 4437–4446.
- Xiong H, Kakei Y, Kobayashi T, et al. 2013a.** Molecular evidence for phytosiderophore-induced improvement of iron nutrition of peanut intercropped with maize in calcareous soil. *Plant, Cell and Environment* **36**: 1888–1902.
- Xiong H, Shen H, Zhang L, et al. 2013b.** Comparative proteomic analysis for assessment of the ecological significance of maize and peanut intercropping. *Journal of Proteomics* **78**: 447–460.
- Xue YF, Yue SC, Zhang YQ, et al. 2012.** Grain and shoot zinc accumulation in winter wheat affected by nitrogen management. *Plant and Soil* **361**: 153–163.
- Xue YF, Eagling T, He JB, et al. 2014a.** Effects of nitrogen on the distribution and chemical speciation of iron and zinc in pearling fractions of wheat grain. *Journal of Agriculture and Food Chemistry* **62**: 4738–4746.
- Xue YF, Yue SC, Zhang W, et al. 2014b.** Zinc, iron, manganese and copper uptake requirement in response to nitrogen supply and the increased grain yield of summer maize. *PloS One* **9**: e93895. <http://dx.doi.org/10.1371/journal.pone.0093895>.
- Xue YF, Zhang W, Liu DY, et al. 2014c.** Effects of nitrogen management on root morphology and zinc translocation from root to shoot of winter wheat in the field. *Field Crops Research* **161**: 38–45.
- Zhang CC, Postma JA, York LM, Lynch JP. 2014.** Root foraging elicits niche complementarity-dependent yield advantage in the ancient ‘three sisters’ (maize/bean/squash) polyculture. *Annals of Botany* **114**: 1719–1733.
- Zhang F, Chen X, Vitousek P. 2013.** Chinese agriculture: an experiment for the world. *Nature* **497**: 33–35.
- Zhang L, Fan J, Ding X, He X, Zhang F, Feng G. 2014.** Hyphosphere interactions between an arbuscular mycorrhizal fungus and a phosphate solubilizing bacterium promote phytate mineralization in soil. *Soil Biology and Biochemistry* **74**: 177–183.
- Zhao FJ, McGrath SP. 2009.** Biofortification and phytoremediation. *Current Opinion in Plant Biology* **12**: 373–380.
- Zhu C, Naqvi S, Gomez-Galera S, Pelacho AM, Capell T, Christou P. 2007.** Transgenic strategies for the nutritional enhancement of plants. *Trends in Plant Science* **12**: 548–555.
- Zhu YG, Smith FA, Smith SE. 2002.** Phosphorus efficiencies and their effects on Zn, Cu, and Mn nutrition of different barley (*Hordeum vulgare*) cultivars grown in sand culture. *Crop and Pasture Science* **53**: 211–216.
- Zuo Y, Li X, Wang Y, Cao Y, Zhang F. 1997.** Effect of maize/peanut intercropping on iron nutrition of peanut. *Plant Nutrition and Fertilizer Science* **3**: 153–159. [in Chinese]
- Zuo Y, Li X, Wang Q, Cao Y, Zhang F. 1998a.** Study on mechanisms of improvement of iron nutrition of peanut by intercropping with maize or wheat. *Acta Ecologica Sinica* **18**: 489–495. [in Chinese]
- Zuo Y, Li X, Zhang F, Cao Y, Wang Y. 1998b.** The effects of maize/peanut intercropping on phytosiderophore secretion of maize root and iron nutrition of peanut. *Journal of Huazhong Agricultural University* **17**: 357–363. [in Chinese]
- Zuo Y, Zhang F, Li X, Cao Y. 2000.** Studies on the improvement in iron nutrition of peanut by intercropping

with maize on a calcareous soil. *Plant and Soil* **220**: 13–25.

Zuo Y, Li X, Cao Y, Zhang F, Christie P. 2003. Iron nutrition of peanut enhanced by mixed cropping with maize: possible role of root morphology and rhizosphere microflora. *Journal of Plant Nutrition* **26**: 2093–2110.

Zuo Y, Zhang F. 2008. Effect of peanut mixed cropping with gramineous species on micronutrient concentrations and iron chlorosis of peanut plants grown in a calcareous soil. *Plant and Soil* **306**: 23–36.

Zuo Y, Zhang F. 2009. Iron and zinc biofortification strategies in dicot plants by intercropping with gramineous species. A review. *Agronomy for Sustainable Development* **29**: 63–71.

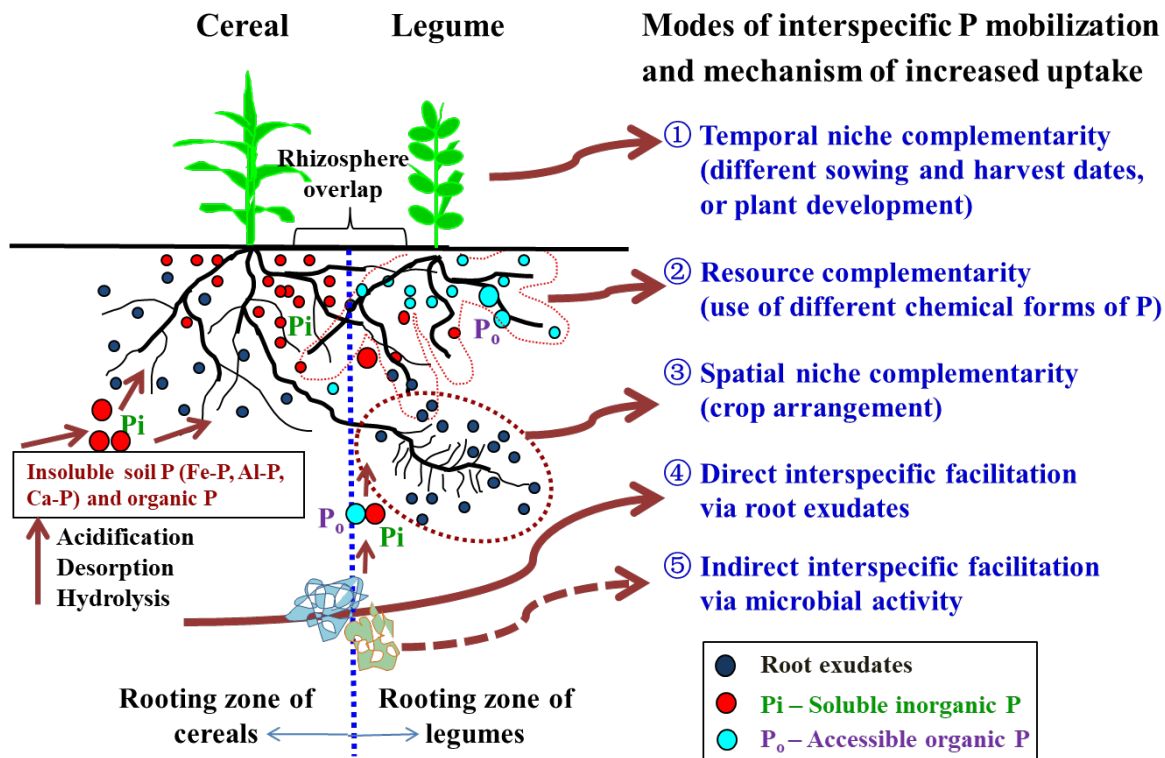


FIG. 1. Schematic overview of possible direct and indirect modes of increased phosphorus (P) uptake in cereal-legume intercropping. Temporal, spatial and resource complementarity and direct and indirect mobilization are indicated.

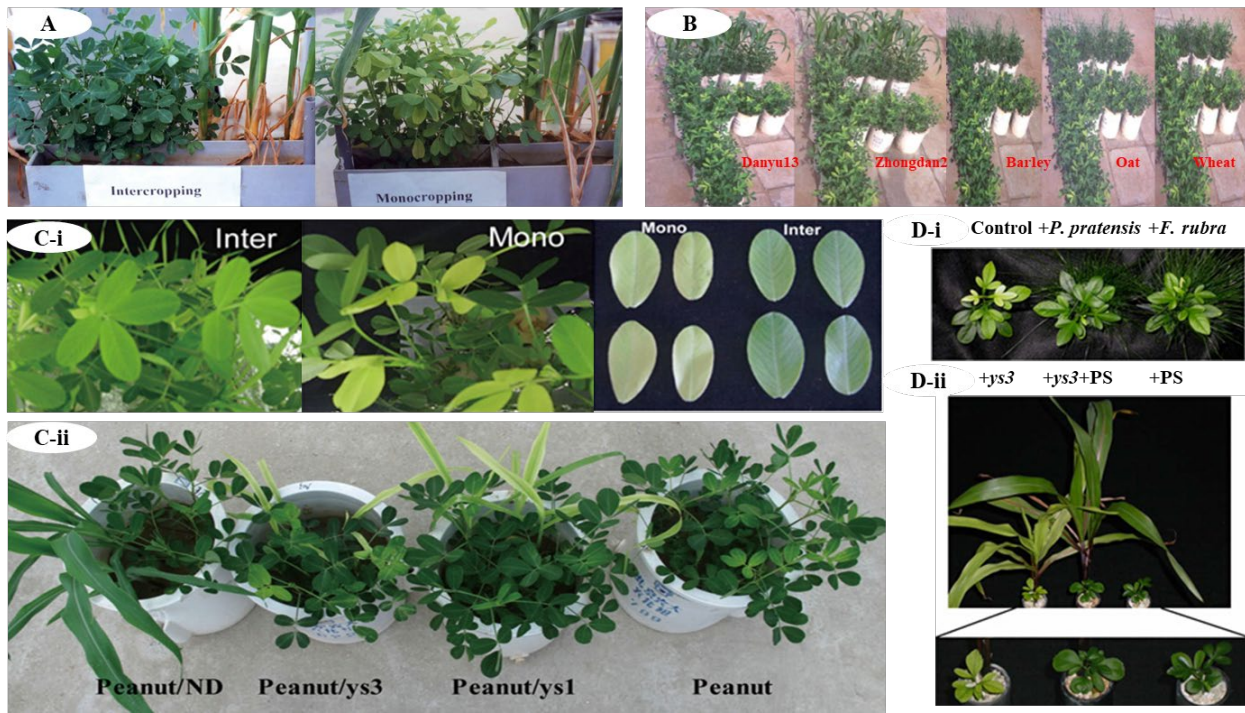


FIG. 2. Images of studies conducted in rhizoboxes (A), pots (B, C-ii and D) and hydroponic cultures (C-i) on alleviation of Fe chlorosis in peanut and citrus by intercropping with graminaceous species. (A) Peanuts intercropped with maize showing no Fe-deficiency symptoms (left) and peanuts without root interaction with maize showing Fe chlorosis (right) in a calcareous sandy soil. (B) Alleviation of Fe chlorosis in young leaves of peanut plants grown together with two maize genotypes (Danyu 13 and Zhongdan 2), barley, oat or wheat in a calcareous sandy soil. (C-i) Improved Fe nutrition in peanuts co-cultured with Fe-deficient wheat (Inter) compared with mono-cultured peanuts (Mono) in hydroponic culture. (C-ii) Alleviation of Fe chlorosis in young leaves of peanut grown with maize of wild type (ND, Nongda 108) or *yellow stripe 1* (*ys1*) mutant unable to take up phytosiderophores (PS) compared to mono-cropped peanut and peanut intercropped with the PS secretion-defective *yellow stripe 3* (*ys3*) mutant in a calcareous soil. (D-i) Alleviation of Fe chlorosis in leaves of Citrumelo 'Swingle' grown for five weeks together with *Poa pratensis* L. or *Festuca rubra* L. in a calcareous soil. (D-ii) 'Swingle' grown in intercropping with *yellow stripe 3* (*ys3*) maize plants unable to release phytosiderophores (PS), showing clear Fe chlorosis, compared to PS-amended plants. (A), (B) and (D) reproduced from Zuo and Zhang (2009), Zuo and Zhang (2008) and Cesco *et al.* (2006), respectively, with the permission of Springer Science and Business Media. (C) reproduced from Xiong *et al.* (2013a) with the permission of John Wiley and Sons.

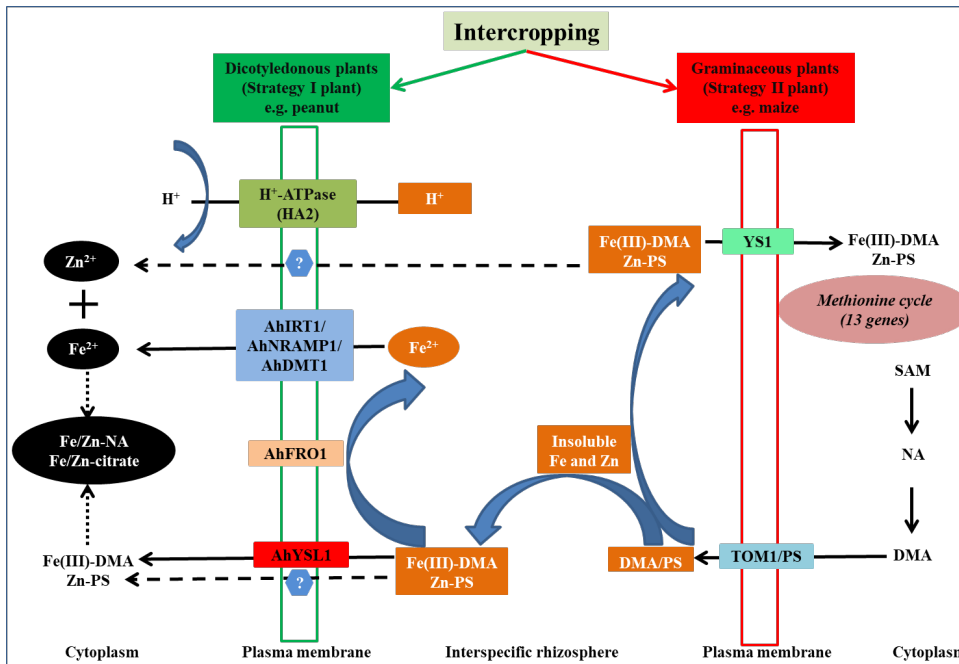


FIG. 3. Model of molecular mechanisms of phytosiderophore (PS)-mediated Fe and Zn uptake in dicotyledonous plants (Strategy I, e.g. peanut) intercropped with graminaceous plants (Strategy II, e.g. maize). Maize plants secrete PS or deoxymugineic acid (DMA) into the rhizosphere via the transporter of mugineic acid 1 (TOM1) or PS. In soil, otherwise insoluble Fe and Zn are desorbed and incorporated into Fe(III)-PS and Zn-PS complexes. The Fe-PS and Zn-PS are taken up by maize via the yellow stripe 1 (YS1) plasma membrane transporter of maize roots. Three molecules of *S*-adenosylmethionine (SAM) supplied from the methionine cycle combine to form nicotianamine (NA) which is converted to DMA for a continuous supply. The Fe(III)-DMA may be utilized by peanut via two pathways: reduction- or transporter-mediated uptake. In the reduction-based pathway, peanut roots release H⁺ through H⁺-ATPase 2 (HA2) and reduces Fe³⁺ via the ferric reduction oxidase 1 (AhFRO1) to Fe²⁺, which is then taken up via iron-regulated transporter 1 (AhIRT1), natural resistance-associated macrophage protein 1 (AhNRAMP1), or divalent metal-ion transporter 1 (AhDMT1). In the transporter-mediated pathway, the Fe(III)-DMA is taken up directly via the yellow stripe1-like 1 (AhYSL1) transporter in the root epidermis. Free Zn²⁺ and Zn-PS may be taken up like Fe. Root-shoot translocation of Fe and Zn is mediated by Fe/Zn-NA or Fe/Zn-citrate complexes (Cornu *et al.*, 2015). ‘?’ indicates unknown pathway. The illustration is based on Varanini *et al.* (2008), Zuo and Zhang (2009) and Xiong *et al.* (2013a), with the permission of John Wiley and Sons.

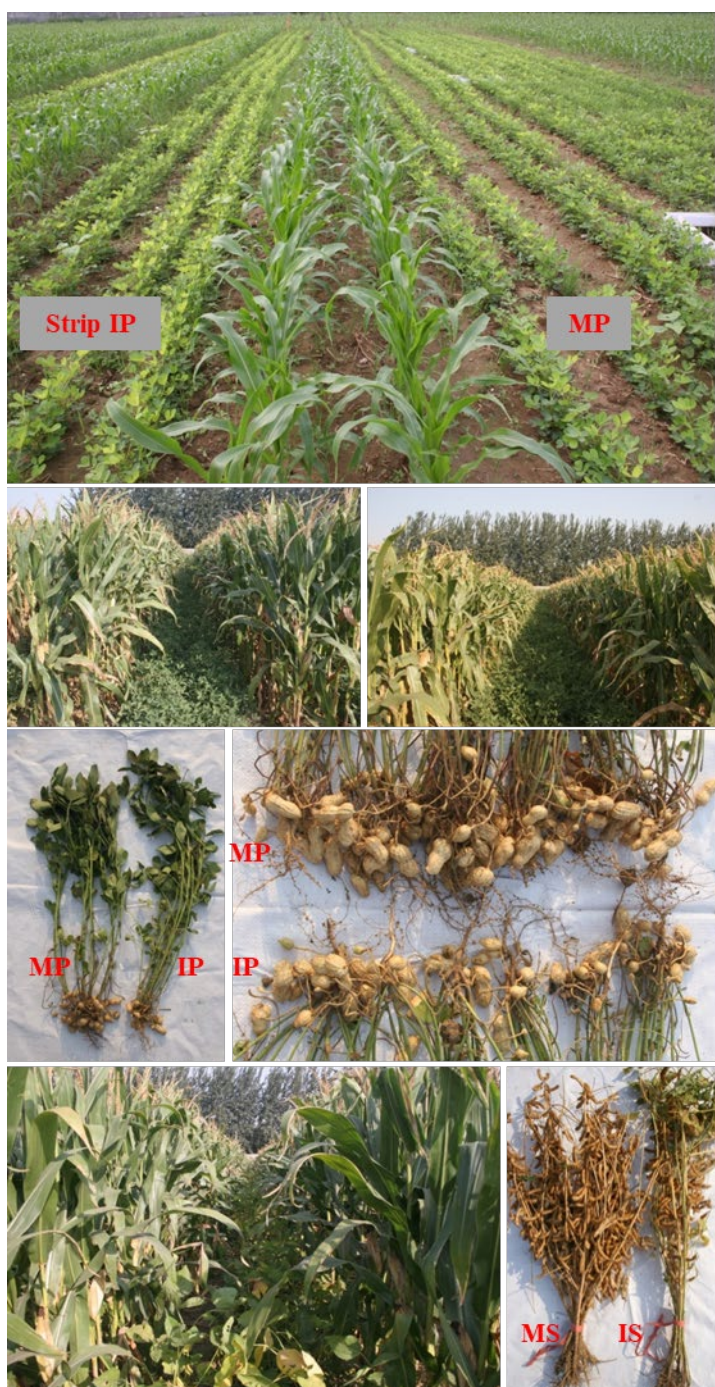


FIG. 4. Peanut and soybean strip intercropping with maize in the field. (A) Strip intercropping unable to correct Fe chlorosis in peanuts (IP) on a poorly-drained calcareous soil compared to mono-cropped peanuts (MP). (B) Two rows of maize intercropped with two rows of peanut. (C) Two rows of maize intercropped with four rows of peanut. (D) Comparison of a single plant of mono-cropped peanut (MP) with a single plant of intercropped peanut (IP) from (B). (E) Comparison of pods of six mono-cropped peanut plants (MP) with six intercropped peanut plants (IP) from (B). (F) Two rows of maize intercropped with two rows of soybean. (G) Comparison of 10 mono-cropped soybean plants (MS) with 10 intercropped soybean plants (IS) from (F).

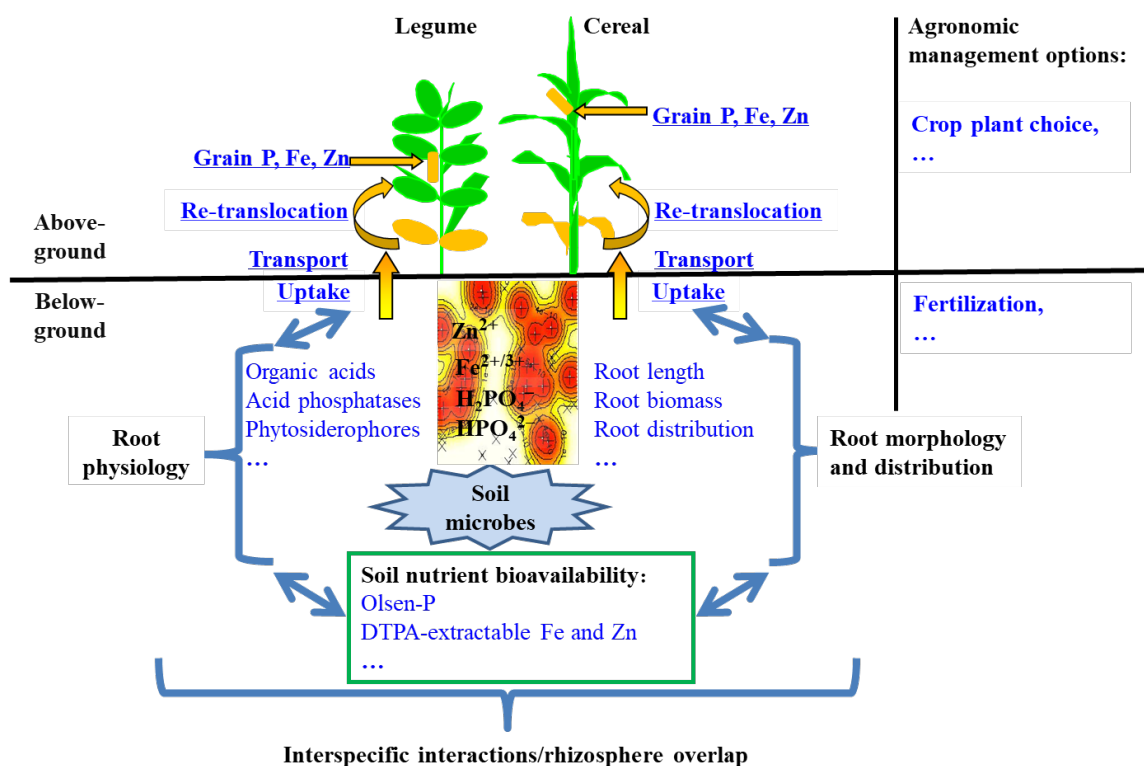


FIG. 5. Summary overview of soil/rhizosphere-plant processes involved in P, Fe and Zn uptake from soil and translocation to grains in legume-cereal intercropping.

TABLE 1. *List of studies showing increased phosphorus (P) uptake in intercropping systems compared to monocropping*

Authors	Crop species combination	Study type	Soil pH	P source	Beneficiary	Loser	Types of improved P uptake	Study of root physiology and morphology	Suggested mechanism
Gardner and Boundy (1983)	Wheat/lupin	Field	Alkaline	Inorganic	Wheat	None	(2)	None	Facilitation
Horst and Waschkiew (1987)	Spring wheat/white lupin	Pot	Acid	Inorganic	Wheat	–	–	Yes	Facilitation
Li <i>et al.</i> (2001a, b)	Wheat/maize or soybean	Field	Alkaline	Inorganic	Wheat	None	(2)	None	Niche differentiation
Dessougi <i>et al.</i> (2003)	Maize/groundnut	Pot	Acid	Inorganic	Maize	–	–	Yes	Increased root length and root P-uptake rate
Li <i>et al.</i> (2003a)	Wheat/chickpea	Pot	Acid	Organic	Wheat	Chickpea	(3)	Only physiology	Facilitation
Li <i>et al.</i> (2003b)	Maize/faba bean	Field	Alkaline	Inorganic	Both	None	(1)	Root barrier	Root interactions
S. Li <i>et al.</i> (2004)	Maize/chickpea	Hydroponic	Acid solution alkaline	Organic	Maize	Chickpea	(3)	Only physiology	Facilitation
Cu <i>et al.</i> (2005)	Wheat/white lupin	Soil column	Alkaline	–	Wheat	None	(2)	Only physiology	Complementarity
Li <i>et al.</i> (2007)	Maize/faba bean	Field	Alkaline	Inorganic	Both	None	(1)	Only physiology	Facilitation
Wang <i>et al.</i> (2007)	Wheat/chickpea	Pot	Acid	Inorganic	Both	None	(1)	Yes	Rhizosphere interactions
Hauggaard-Nielsen <i>et al.</i> (2009)	Barley/spring pea	Field	Acid	–	Barley	Pea	(3)	None	None
Li <i>et al.</i> (2009)	Rice/mung bean with AM inoculation	Pot	Acid	Inorganic	Both	None	(1)	Only morphology	Improved AM colonisation
Betencourt <i>et al.</i> (2012)	Durum wheat/chickpea	Pot	Alkaline	Inorganic	Both	None	(1)	Only physiology	Facilitation
Mei <i>et al.</i> (2012)	Maize/faba bean	Field	Alkaline	Inorganic	Both	None	(1)	None	Complementarity and facilitation
Xia <i>et al.</i> (2013a)	Maize/faba bean, chickpea, soybean or turnip	Field	Alkaline	Inorganic	Maize, faba bean, chickpea.	Soybean	(1) + (3)	None	Complementarity and facilitation
H. Li <i>et al.</i> (2014)	Maize/faba bean	Rhizobox	Alkaline	Inorganic	Faba bean	None	(2)	Only physiology	Facilitation
Lelei and Onwonga (2014)	Maize/white lupin	Field	Neutral	Inorganic	Maize	Lupin	(3)	None	Facilitation
Dissanayaka <i>et al.</i> (2015)	Maize/white lupin	Pot	Acid	Inorganic	Maize	None	(2)	Yes	Facilitation

–, data not available; AM, arbuscular mycorrhizas.

Type of improved P uptake: (1), Both partners benefit; (2), One partner benefits with no negative effect on its companion; (3), One partner benefits with negative effect on its companion.

Wheat (*Triticum aestivum* L.), white lupin (*Lupinus albus* L.), maize (*Zea mays* L.), soybean (*Glycine max* L.), groundnut or peanut (*Arachis hypogaea* L.), chickpea (*Cicer arietinum* L.), faba bean (*Vicia faba* L.), barley (*Hordeum vulgare* L.), spring pea (*Pisum sativum* L.), rice (*Oryza sativa* L.), mung bean (*Vigna radiata* L.), turnip (*Brassica campestris* L.).

TABLE 2. *Growth and Fe nutritional responses of dicotyledonous plants to intercropping with graminaceous plants and proposed mechanisms of chlorosis alleviation*

Authors	Crop species combination	Study type	pH of growth medium	Growth stage	Root Fe conc.	Young leaf chlorophyll conc.	Young leaf active Fe conc.	Shoot /stem Fe conc.	Seed Fe conc.	Root biomass	Shoot biomass	Seed biomass	Total biomass	Study of root morphology & physiology	Suggested mechanism
Zuo <i>et al.</i> (1997)	Maize/peanut	Rhizobox	Alkaline	3 months	+	+	+	+	+	+	-	+	+	Only morphology	Physiology
Zuo <i>et al.</i> (1998a)	Maize/peanut	Pot	Alkaline	2 months	+	+	+	+	N/A	+	+	N/A	+	Yes	Physiology
	Wheat/peanut	Pot	Alkaline	2 months	+	+	+	+	N/A	+	+	N/A	+	Yes	Physiology
Zuo <i>et al.</i> (1998b)	Maize/peanut	Soil-solution	Alkaline-acid	N/A	N/A	N/A	+	N/A	N/A	ns	ns	N/A	N/A	Yes	Physiology Rhizosphere
Zuo <i>et al.</i> (2000)	Maize/peanut	Field	Alkaline	Flowering	N/A	+	+	N/A	N/A	+	-	N/A	N/A	No	effect of maize
Fang <i>et al.</i> (2004)	Maize/peanut	Pot	Alkaline	Acicula	ns	N/A	+	+	N/A	N/A	N/A	N/A	+	Soil microbes	Root interactions Physiology, isotope
Cesco <i>et al.</i> (2006)	Poa or F/C	Pot	Alkaline	5 weeks	N/A	+	N/A	N/A	N/A	N/A	N/A	N/A	N/A	method	Direct facilitation
Inal <i>et al.</i> (2007)	Maize/peanut	Pot	Alkaline	35–60 days	N/A	+	N/A	+	N/A	N/A	-	N/A	N/A	Only physiology	Physiology
Zuo and	Maize, barley														
Zhang (2008)	oat or wheat/peanut	Pot	Alkaline	55–70 days	N/A	+	+	+	N/A	+	ns	N/A	N/A	Only physiology	Physiology
Ding <i>et al.</i> (2009)	Maize/peanut	Pot	Alkaline	Flowering	N/A	+	N/A	N/A	N/A	N/A	N/A	N/A	N/A	Only physiology	Physiology
Jiao <i>et al.</i> (2012)	Maize/peanut	Field	Alkaline	Maturity	N/A	N/A	N/A	+	+	N/A	-	N/A	N/A	No	Physiology
Xiong <i>et al.</i> (2013a)	Maize/peanut	Pot	Alkaline	32–35 days	N/A	+	+	N/A	N/A	N/A	N/A	N/A	N/A	Only physiology,	Direct interspecific
	Wheat/peanut	Hydroponic	Alkaline	2 weeks	+	+	N/A	N/A	N/A	N/A	N/A	N/A	N/A	molecular method	facilitation
Xiong <i>et al.</i> (2013b)	Maize/peanut	Pot	Alkaline	63 days	N/A	+	+	N/A	N/A	N/A	N/A	N/A	ns	Only physiology	Physiology
Shen <i>et al.</i> (2014)	Maize/peanut	Pot	Alkaline	45 days	N/A	N/A	ns	N/A	N/A	N/A	N/A	N/A	ns		Physiology
				65 days	N/A	N/A	+	N/A	N/A	N/A	N/A	N/A	ns	Only physiology	

Crop species: barley (*Hordeum vulgare* L.); C, citrumelo ‘Swingle’; F, *Festuca rubra* L.; maize (*Zea mays* L.); oat (*Avena sativa* L.); Poa, *Poa pratensis* L.; peanut (*Arachis hypogaea* L.); wheat (*Triticum aestivum* L.).

conc., concentration; +, increase; -, decrease; ns, not significant; N/A, data not available.