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REGULAR ARTICLE

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

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Abstract
Aims This study of a maize-white lupin model cropping system was conducted to investigate the effects of rhizosphere-sharing of white lupin, a P-efficient plant, on growth and P accumulation of maize under different P rates and forms in two contrasting soils.
Methods With Regosol and Andosol, a 42-day pot experiment was conducted for 0P (no P addition), 50Pi, 100Pi (50 and 100 mg P kg^{-1} soil by $NaHPO_4 \cdot 2H_2O$ respectively), and 100Po (100 mg P kg^{-1} soil by phytate). Plant growth, P uptake, rhizosphere pH, and different P fractions were investigated.
Results Complementary effects of intercropping for maize were observed in Regosol, but not in Andosol. Total P uptake by intercropped maize in 0P, 50Pi, and 100Po was elevated by 46, 37, and 65 %, respectively, compared to when it was grown as a monoculture. White lupin mobilized P from sparingly soluble forms. Thereby, maize plant enhanced its P accumulation as a result of access to these two fractions in mixed culture in Regosol, where strong root intermingling occurred among intercropped plants.
Conclusions Results suggest that the P mobilization strategy of white lupin from sparingly soluble P pools in soil can enhance the P acquisition efficiency of coexisting maize with P facilitation in this intercropping occurring in the direction of white lupin to maize. Achieving enhanced growth and P uptake by P-inefficient species in intercropping with white lupin is dependent on the type of soil in which those plants are grown.

Keywords Intercropping · Rhizosphere-sharing · P facilitation · P availability · Soil P pools · Soil type

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Introduction

Although phosphorus (P) plays a vital role in the energy metabolism and in biosynthesis of nucleic acid and membranes, it is regarded as the second most frequently limiting major nutrient for plant growth (Raghothama 1999). In most soils, the P concentration in soil solution is inadequate for optimal growth of many crop plants (Hinsinger 2001; Raghothama 1999). Consequently, P-deficiency is a major yield-limiting factor for plants, especially in acidic and calcareous soils, where P retention is high (Hinsinger 2001). In this context, it is necessary to apply P fertilizers continually to sustain crop production. However, increasing the fertilizer P usage further is not an option to improve agricultural production to meet the global food demand (Hinsinger et al. 2011; Vance 2001) because phosphate rocks that are used to manufacture fertilizer P are a finite resource

(Cordell et al. 2009; Dawson and Hilton 2011) that is expected to be exhausted by the end of the century (Gilbert 2009). It is therefore important to use alternatives such as P-efficient genotypes and alternative management strategies to exploit soil P resources better through increasing P bioavailability in agroecosystems (Lambers et al. 2006; Vance 2001).

Richardson et al. (2011) reported three strategies for plants to increase production in P-deficient soils and to reduce the amounts of P fertilizers that are necessary to enhance production: (i) 'root foraging strategies', which enable plant to explore large volumes of soil and thereby acquire more P from soil; (ii) 'P mining strategies', which increase desorption and mineralization of sparingly available P pools and organic P pools in soil by root exudates, organic anions, and phosphatases; and (iii) 'improved internal P-utilization efficiency', which might be helpful for plants to produce a higher yield per unit of P uptake. Plants that are able to use any of these strategies would use soil P and fertilizer P efficiently, irrespective of the application of fertilizer P to soil (Simpson et al. 2011).

Several legume species that exude organic anion to access sparingly available P have been examined. Those crop species include white lupin (Cu et al. 2005; Gardner and Boundy 1983; Hocking and Randall 2001), pigeon pea (*Cajanus cajan* L. Millsp.; Ae et al. 1990), faba bean (*Vicia faba* L.; Li et al. 2007), and chickpea (*Cicer arietinum* L.; Veneklaas et al. 2003). Traits of some lupins, such as formation of cluster roots and exudation of vast amounts of phosphate-mobilizing substances, have made them ideal plants to thrive in soils where higher amounts of P remain in poorly available forms for most plants (Lambers et al. 2013). White lupin has these desirable traits to a greater degree. Formation of cluster roots (proteoid roots) in response to low P supply (Gardner et al. 1981; Keerthisinghe et al. 1998) and exudation of organic acids and phosphatase from these specific root structures have been demonstrated in earlier studies (Dinkelaker et al. 1989; Gardner et al. 1983). Organic anion exudation into the rhizosphere increases the mobilization of sparingly soluble soil P (Gardner et al. 1983; Gerke et al. 1994; Li et al. 1997), whereas phosphatase secretion by white lupin helps it to use organic P fractions in addition to inorganic P in soil (Adams and Pate 1992; Lambers et al. 1998). Enhanced ability of white lupin to secrete acid phosphatase under P-deficient conditions has also been described by Tadano and Sakai (1991) and by Wasaki et al. (2003). Lupins can mobilize sparingly available nutrients, mainly P and micronutrients, not only for themselves but also for interplanted or subsequent crops (Lambers et al. 2013). Therefore, the ability of white lupin to secrete these types of P-mobilizing substances and to access more recalcitrant sources of P in soil highlights its potential applicability to improve P inefficient species through sharing of rhizosphere functions in mixed cultures.

For a particular crop plant, P benefit can be achieved by integrating plant species that use sparingly available phosphate and organic P in intercropping systems. In this context, P-mobilizing species can enhance the growth and P uptake by companion cereals (Simpson et al. 2011). Enhanced P acquisition and growth have been demonstrated previously for different cereals in mixed culture systems with legumes. Evidence of positive responses by interplanted cereals with legumes includes that of wheat intercropped with chickpea (Li et al. 2003), white lupin (Cu et al. 2005; Kamh et al. 1999), and faba bean (Song et al. 2007). Other cereals include maize (Li et al. 2004) and barley (Gunes et al. 2007) intercropped with chickpea and sorghum intercropped with pigeon pea (Ae et al. 1990).

In cereal legume intercropping systems, the P acquisition of cereal is enhanced because of the legumes' ability to exude large amounts of P-mobilizing compounds, such as carboxylates, that can mobilize sparingly available P (Neumann and Römheld 1999; Pearse et al. 2006; Veneklaas et al. 2003), phosphatases that can mineralize organic P (Nuruzzaman et al. 2006), and protons from N_2 fixation (Hinsinger et al. 2003; Tang et al. 1997) that ultimately increase P availability. Soil pH is known to be a crucially important parameter determining the P availability in soil (Hinsinger 2001). Acidification of the rhizosphere is expected in cereal–legume mixed cultures as a result of proton release by roots of N_2-fixing legumes (Cu et al. 2005; Li et al. 2008). This acidification can be expected to benefit plants grown in alkaline soils. However, few studies to date have examined the potential role of rhizosphere pH in intercropping systems.

Most studies of P mobilization for a main crop plant by a P-efficient plant species in mixed cultures have been confined to soils with low P availability because the P mobilization capacity is enhanced under P-deficient conditions (Neumann et al. 1999). Crop cultivators apply fertilizer P to cropping lands to reduce the risk of P-deficiency during crop growth either by organic or inorganic fertilizer sources. To date, positive effects of intercropping have been reported mainly in alkaline soils. Facilitation of P uptake in the intercropping of P-inefficient and efficient plant species under different rates and forms of P supply have been found only in a few studies. Given these circumstances, the P uptake of main crop plants from different P pools as influenced by interplanted P-efficient plant species has yet to be investigated. Consequently, this study addressed this knowledge gap by assessing the growth and P-advantages of maize as main crop plant intercropped with white lupin as P-efficient plant under different P rates and forms in two contrasting acidic soils. This study also evaluated the various soil P fractions in the rhizosphere of monocropped and intercropped plants species. Our study fundamentally examined the early stage growth of the two plants. White lupin was selected as the P-efficient plant species for this study because of its enhanced ability to secrete P mobilizing substances, by which it accesses less-labile P pools in soil, which are not available to other plants.

Material and methods

Soils

Two soil types, Regosol and Andosol, were collected respectively from a field at Hiroshima University at Higashi-Hiroshima and a forest at Shobara, Hiroshima, Japan. Soil was air-dried and passed through a 2-mm sieve. Table 1 presents physical and chemical parameters of the soils. Both soils were acidic with contrasting P adsorption capacities.

Table 1 Physical and chemical properties of two soils used in the experiment. Data represent the mean of three replicates±SE (standard errors)

Soil property	Regosol	Andosol
Bulk density (g cm^{-3})	1.26±0.01	0.72±0.02
Water holding capacity (mL 100 g^{-1})	43.81±3.82	99.17±3.76
Total carbon (g kg^{-1})	24.92±0.09	156.64±0.35
Total nitrogen (g kg^{-1})	1.01±0.01	5.43±0.02
pH (1:2.5 soil:H_2O)	5.40±0.01	5.11±0.03
P adsorption capacity (g-P_2O_5 100 g^{-1})	0.17±0.00	1.98±0.01

Plant growth conditions

A pot experiment was conducted in a naturally lit glass house at Hiroshima University. The two plant species examined were white lupin (*Lupinus albus* L. cv. Energy) and maize (*Zea mays* L. cv. Snow-dent 125 'Wakaba') either as monocropping or mixed cropping. Four P fertilizer treatments were no P addition (0P), 50 mg P kg^{-1} soil as $NaHPO_4{\cdot}2H_2O$ (50Pi), 100 mg P kg^{-1} soil as $NaHPO_4{\cdot}2H_2O$ (100Pi), and 100 mg P kg^{-1} soil as phytic acid, dodecasodiumsalt (100Po). Regosol was mixed with 10 % (w/w) peat moss. Then, to adjust the pH, $CaCO_3$ was added to a rate of 2 g kg^{-1} of soil because the addition of peat moss can lower pH in Regosol. The two soils were mixed carefully with relevant P rates either as inorganic P or organic P. Both soils were supplied with 100 mg N kg^{-1} soil as NH_4NO_3 and 100 mg K kg^{-1} soil as K_2SO_4. Soil moisture was adjusted to 70 % of field capacity. Pots were filled with 1 kg of prepared soil. Seeds of white lupin and maize were surface-sterilized using 70 % ethanol and were soaked in a polystyrene box under running tap water over 48 h. After radicle emergence, four evenly sized seeds per pot were planted in a monocropping system. In mixed culture, 2:2 combinations from either species were planted per pot. After germination, seedlings were thinned to leave two plants per pot in a monoculture system and a 1:1 combination in mixed culture. Pots were arranged in complete randomized design with three replicates. The arrangements were re-randomized on every seventh days. The experiment also included a pot of unplanted control soil for each P supply for both soils. These pots were examined under the same conditions and were kept for the same duration as the cultivated pots. Soil moisture was maintained at 70 % field capacity by adding deionized water by weight every day. Plants were grown for 42 days.

Plant and soil analysis

At harvest, plants were lifted carefully out of the soil. The root system was shaken gently to remove loosely adhering soil. Then the rhizosphere soil was collected by vigorous shaking of the root system, followed by gentle brushing without damaging the root system. For mixed cropping pots, the root systems of the two species were separated carefully to avoid excessive breakage. Then rhizosphere soil was collected separately for the two species. For sole cropping pots, we collected the rhizosphere soil of two plants together. Immediately after the collection, rhizosphere soil was sieved through 2-mm holes to remove roots. It was then stored at −20 °C until analysis. Shoots and roots were separated after washing the root system with water. The cluster roots formed by white lupin were counted. Then both roots and shoots were oven-dried at 70 °C for 3 days. The dry weights of shoots and roots were recorded to estimate the plant growth. All dried roots and shoots were then ground. A sample of approximately 50 mg was digested using H_2SO_4-H_2O_2. The phosphorus concentration in the digested solution was quantified using vanadomolybdate blue method (Murphy and Reley 1962).

For the determination of soil P pools, sequential fractionation was conducted using the method described by Hedley et al. (1982). Briefly, 0.5 g of rhizosphere and unplanted control soil was weighed and extracted sequentially by shaking overnight (16 h) with a solution. (1) First, 30 mL deionized water was used with two resin strips (Selemion™ ion exchange membrane; Asahi Glass Co. Ltd., Japan). Then P in these resins was extracted by shaking with 20 mL of 0.5 M HCl for 2 h (resin P). (2) Second, 30 mL of 0.5 M $NaHCO_3$ was used after adjustment to pH 8.5 ($NaHCO_3$–P). (3) Subsequently, 30 mL of 0.1 M NaOH (NaOH–P) was used, followed by (4) 20 mL of 1 M HCl (HCl–P). The inorganic P (Pi) concentration in all extracts was determined using the molybdenum blue method (Murphy and Reley 1962). The total P (Pt) in 0.5 M $NaHCO_3$ and 0.1 M NaOH fractions were measured by digestion of the extract with 10 mL of 1.8 N H_2SO_4, and respectively with 0.5 and 0.6 g of $(NH_4)_2S_2O_8$. Organic P (Po) in these fractions was calculated by subtracting the Pi from Pt. Total P of rhizosphere soil was determined separately using vanadomolybdate blue method after digestion of the sample with H_2SO_4-H_2O_2. Residual P was measured as the difference between total P and the sum of all organic and inorganic P fractions. The P adsorption capacity was determined according to the method by Sekiya (1970). Briefly, 5 g of air-dried soil was mixed with 10 mL of 5.869 g-P L^{-1}solutionand was kept for 24 h. Then the solution was centrifuged (8000×g, 1 min) and filtered. The P concentration of filtered solution was measured using molybdenum yellow color method. The P adsorption capacity of each soil type was calculated based on the difference between the P concentration of the original solution added to the soil and the filtered solution. The pH of the rhizosphere soil and unplanted control soil was determined in a water suspension (soil water ratio of 1:2.5) after shaking for one hour.

Statistical analysis

All analyses were conducted using software (SAS ver. 9.1; SAS Institute Inc., Cary, NC, USA). Biomass of shoot, root and whole plant, P concentration of shoot and root, and plant P content data were subjected to two-way ANOVA using the P supply (0P, 50Pi, 100Pi, and 100Po) and cropping system (monocrop and intercrop) as the treatment effects for each crop. This analysis was done separately for each soil. Rhizosphere pH and all P fraction data were subjected to one-way ANOVA with cropping treatments including unplanted control soil (UP, unplanted control soil; MM, monocropped maize; IM, intercropped maize; ML, monocropped lupin; and IL, intercropped lupin) for each P supply under each soil. Significance of the difference between means was assessed using Tukey's Studentized Range Test at the 0.05 probability level. Means are presented with the standard error.

Results

Plant growth and P uptake

This study investigated the rhizosphere-sharing of white lupin, a P-efficient plant, on growth and P acquisition of intercropped maize under different P rates and forms in two soils with contrasting P dynamics. Biomass and P accumulation of both maize and white lupin differed between the two soil types and the P rates used for the study (Tables 2 and 3). Both types of plants recorded higher growth and P uptake in Regosol than in Andosol. For both maize and white lupin, P supply had a significant effect on all plant parameters measured in both soils (Tables 2 and 3). White lupin responded strongly to added phytate in Regosol (Table 2). Neither maize nor white lupin responded to added phytate in Andosol (Table 3). Shoot and root P concentrations were higher in white lupin than in maize under 0P and 100Po in both

Table 2 Dry weight (shoot, root and whole plant), P concentration (shoot and root) and plant P content of maize and white lupin in monocropping and intercropping systems under application of different P rates and forms in Regosol. Values represent the mean of three replicates±SE (standard error)

	P supply	Cropping	Dry weight (g)			P concentration (mg-P/g DW)		Plant P content (mg/plant)
			Shoot	Root	Whole plant	Shoot	Root	
Maize	0P	Monocrop	0.853±0.051 e	0.400±0.047 d	1.253±0.116 e	2.08±0.05 e	1.32±0.04 d	2.30±0.14 e
		Intercrop	0.973±0.064 de	0.433±0.066 cd	1.403±0.091 de	2.84±0.24 cd	1.43±0.05 cd	3.37±0.18 d
	50Pi	Monocrop	1.116±0.048 bc	0.556±0.042 bc	1.673±0.097 bc	3.37±0.08 c	1.76±0.09 c	4.74±0.07 c
		Intercrop	1.253±0.117 b	0.583±0.075 b	1.836±0.131 b	4.08±0.10 b	2.35±0.08 b	6.49±0.31 b
	100Pi	Monocrop	1.446±0.140 a	0.833±0.131 a	2.280±0.118 a	5.02±0.07 a	3.44±0.13 a	10.13±0.29 a
		Intercrop	1.500±0.181 a	0.863±0.161 a	2.363±0.233 a	5.12±0.10 a	3.67±0.13 a	10.84±0.12 a
	100Po	Monocrop	0.873±0.092 e	0.453±0.156 bcd	1.326±0.217 e	2.18±0.04 de	1.42±0.11 cd	2.55±0.18 e
		Intercrop	1.066±0.072 cd	0.486±0.081 bcd	1.546±0.107 cd	3.19±0.18 c	1.71±0.04 c	4.22±0.11 c
	P supply		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
	Cropping		<0.001	0.121	<0.001	<0.001	<0.001	<0.001
	P supply × Cropping		0.174	0.999	0.318	0.018	0.029	0.007
Lupin	0P	Monocrop	0.750±0.041 c	0.147±0.044 c	0.897±0.080 c	3.13±0.09 c	3.15±0.19 cd	2.81±0.07 cd
		Intercrop	0.729±0.081 c	0.144±0.041 c	0.874±0.071 c	3.10±0.24 c	2.96±0.45 d	2.68±0.17 d
	50Pi	Monocrop	0.909±0.056 abc	0.180±0.053 ab	1.090±0.091 abc	4.27±0.40 ab	4.54±0.16 ab	4.72±0.45 b
		Intercrop	0.890±0.083 abc	0.178±0.094 ab	1.069±0.182 abc	4.21±0.23 ab	4.43±0.20 abc	4.55±0.44 b
	100Pi	Monocrop	1.034±0.131 ab	0.193±0.076 a	1.228±0.160 ab	4.89±0.21 a	4.92±0.25 ab	6.02±0.59 a
		Intercrop	1.109±0.192 a	0.195±0.061 a	1.304±0.144 a	4.98±0.28 a	5.17±0.34 a	6.51±0.54 a
	100Po	Monocrop	0.843±0.118 bc	0.166±0.065 abc	1.009±0.161 bc	3.92±0.10 bc	3.84±0.20 bcd	3.94±0.18 bc
		Intercrop	0.830±0.101 bc	0.161±0.040 bc	0.991±0.065 bc	3.86±0.04 bc	3.64±0.16 bcd	3.79±0.32 bcd
	P supply		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
	Cropping		0.885	0.526	0.948	0.917	0.745	0.954
	P supply × Cropping		0.768	0.992	0.785	0.984	0.821	0.683

P values in the table are related with the two-way analysis of variance for the factors P supply, cropping and the interaction of P supply × cropping

0P no P addition, *50Pi* 50 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Pi* 100 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Po* 100 mg P kg^{-1} soil by phytate

Values in a column not followed by a common letter are significantly different ($P<0.05$)

soils. However, maize plants were somewhat larger than white lupin in each P supply (Tables 2 and 3). Positive effects of intercropped white lupin on growth and P accumulation of maize were observed only in Regosol where mixed culture increased dry weight of shoot and whole plant, P concentration of root and shoot, and plant P content of maize. This effect was significant in 0P, 50Pi, and 100Po treatments (Table 2). The respective plant P contents of intercropped maize in 0P, 50Pi, and 100Po were 46, 37, and 65 % greater than those in monoculture systems in Regosol. In the same soil for the same P dosages, the plant dry matter increments were, respectively, 12, 10, and 17 % (Table 2). In Andosol, cropping had no effect on maize (Table 3).

Cluster root formation by white lupin and rhizosphere pH changes in two soils

White lupin produced cluster roots irrespective of the soil type and addition of soluble or organic P sources except for the 100Pi supply in Regosol (Fig. 1). Cluster root formation was much more pronounced in Andosol than in Regosol. Each soil type exhibited a tendency to decrease the cluster root formation with increasing added soluble P dosage. Under 100Pi treatment, cluster root formation was suppressed completely in Regosol, but not in Andosol (Fig. 1).

Irrespective of the soil type and cropping system, white lupin decreased its rhizosphere pH markedly under all P rates (Table 4). Reduction of rhizosphere pH of

Table 3 Dry weight (shoot, root and whole plant), P concentration (shoot and root) and plant P content of maize and white lupin in monocropping and intercropping systems under application of different P rates and forms in Andosol. Values represent the mean of three replicates±SE (standard errors)

	P supply	Cropping	Dry weight (g)			P concentration (mg-P/g DW)		Plant P content (mg/plant)
			Shoot	Root	Whole plant	Shoot	Root	
Maize	0P	Monocrop	0.493±0.069 b	0.203±0.101 c	0.700±0.072 c	1.40±0.15 c	1.02±0.18 b	0.90±0.03 c
		Intercrop	0.500±0.041 b	0.213±0.087 bc	0.713±0.107 c	1.42±0.13 c	1.10±0.29 b	0.96±0.14 c
	50Pi	Monocrop	0.710±0.097 a	0.323±0.074 ab	1.036±0.131 b	2.21±0.14 b	1.69±0.21 ab	2.12±0.07 b
		Intercrop	0.733±0.103 a	0.326±0.079 ab	1.060±0.185 ab	2.28±0.14 b	1.71±0.32 ab	2.23±0.16 b
	100Pi	Monocrop	0.806±0.120 a	0.396±0.081 a	1.206±0.162 ab	2.94±0.17 a	2.03±0.28 a	3.18±0.18 a
		Intercrop	0.820±0.111 a	0.413±0.113 a	1.230±0.133 a	3.04±0.27 a	2.16±0.06 a	3.38±0.12 a
	100Po	Monocrop	0.506±0.075 b	0.210±0.045 bc	0.713±0.106 c	1.48±0.05 c	1.10±0.07 b	0.97±0.09 c
		Intercrop	0.513±0.054 b	0.230±0.047 bc	0.743±0.080 c	1.51±0.12 c	1.19±0.17 b	1.05±0.12 c
	P supply		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
	Cropping		0.564	0.441	0.424	0.534	0.502	0.171
	P supply × Cropping		0.991	0.982	0.997	0.987	0.990	0.915
Lupin	0P	Monocrop	0.480±0.049 e	0.120±0.041 bc	0.601±0.072 e	2.40±0.19 bc	2.28±0.14 ab	1.43±0.10 cd
		Intercrop	0.468±0.052 e	0.114±0.061 c	0.583±0.076 e	2.23±0.08 c	2.09±0.26 b	1.29±0.15 d
	50Pi	Monocrop	0.564±0.083 bc	0.141±0.052 abc	0.705±0.090 bc	3.17±0.08 abc	2.94±0.06 ab	2.19±0.13 bc
		Intercrop	0.556±0.071 cd	0.136±0.067 abc	0.692±0.064 cd	3.02±0.09 abc	2.83±0.14 ab	2.08±0.22 bc
	100Pi	Monocrop	0.649±0.072 a	0.156±0.043 a	0.806±0.080 a	3.81±0.08 a	3.31±0.30 a	2.99±0.17 a
		Intercrop	0.629±0.109 ab	0.154±0.046 ab	0.784±0.083 ab	3.68±0.13 ab	3.26±0.35 a	2.82±0.15 ab
	100Po	Monocrop	0.487±0.098 de	0.124±0.059 abc	0.611±0.067 de	2.48±0.10 bc	2.31±0.20 ab	1.49±0.06 cd
		Intercrop	0.481±0.051 e	0.121±0.081 bc	0.602±0.052 de	2.42±0.14 bc	2.26±0.12 ab	1.44±0.06 cd
	P supply		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
	Cropping		0.249	0.465	0.249	0.479	0.532	0.251
	P supply × Cropping		0.954	0.995	0.983	0.997	0.987	0.975

P values in the table are related with the two-way analysis of variance for the factors P supply, cropping and the interaction of P supply × cropping

0P no P addition, *50Pi* 50 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Pi* 100 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Po* 100 mg P kg^{-1} soil by phytate

Values in a column not followed by a common letter are significantly different ($P<0.05$)

white lupin was low with the application of 100Pi as soluble P compared to no P addition, 50Pi, and 100Po dosages, where this effect was observed especially more in Regosol than in Andosol (Table 4). In the case of maize, no significant change of pH was observed in the rhizosphere of a monoculture system compared to bulk soil. Incorporation of white lupin as a P-efficient companion plant into the maize cropping system lowered the pH in the maize rhizosphere compared to sole maize cropping. The reduction of pH in intercropped maize rhizosphere was intermediate between monocropped maize and monocropped or intercropped white lupin (Table 4). It is noteworthy that this effect was pronounced in Regosol, but in Andosol, rhizosphere pH reduction of intercropped maize was negligible (Table 4).

Rhizosphere P fractions

Compared to a lack of P addition, the addition of soluble P as $NaH_2PO_4{\bullet}2H_2O$ increased the inorganic pools, particularly those of resin P and $NaHCO_3$–Pi, both in Regosol and Andosol (Tables 5 and 6), although it increased NaOH–Pi only in Andosol (Table 6). The application of phytate as an organic P source increased mainly NaOH–Po and residual P in both soils (Tables 5 and 6), although it produced a shift in $NaHCO_3$–Po only in Regosol (Table 5). The increment of resin P with the

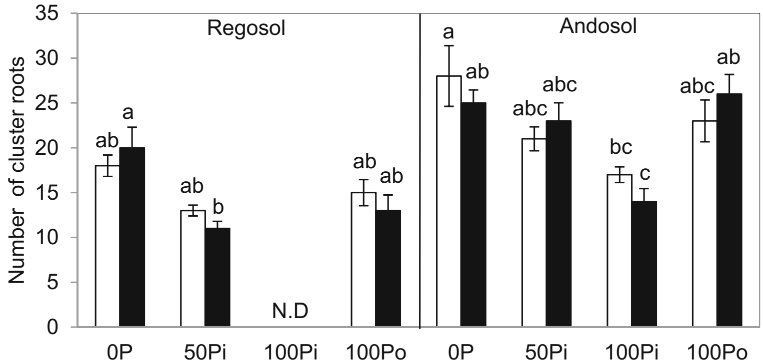

Fig. 1 Cluster root formation by white lupin under different rates and forms of P in two soils. *Black columns* represent white lupin in an intercropping system with maize, whereas others show data for sole cropping. *Different letters* denote significant differences between treatments ($P \leq 0.05$) by Tukey's Studentized Range Test. *Error bars* represent the standard error (S.E.; $n=3$). *0P* no P addition; *50Pi* 50 mg P kg^{-1} soil by $NaH_2PO_4 \cdot 2H_2O$; *100Pi* 100 mg P kg^{-1} soil by $NaH_2PO_4 \cdot 2H_2O$; *100Po* 100 mg P kg^{-1} soil by phytate. (N.D: Cluster root formation was not observed under *100Pi* treatment in Regosol)

addition of soluble P was greater in Regosol than in Andosol (Tables 5 and 6).

Inorganic labile pools, resin P, and $NaHCO_3$–Pi were significantly lower in the rhizosphere of maize and white lupin in both soils, irrespective of the cropping system, with the stronger reduction in maize rhizosphere rather than in white lupin rhizosphere (Tables 5 and 6). $NaHCO_3$–Po, which is regarded as a labile pool, was significantly lower in the maize rhizosphere in both soils (Tables 5 and 6). Surprisingly, this labile organic P pool was augmented in the white lupin rhizosphere in Regosol (Table 5), although its concentration was lower in Andosol (Table 6).

No apparent change of NaOH–Pi and NaOH–Po fractions was noted in maize rhizosphere alone in any soil type (Tables 5 and 6). In contrast, concentration of these sparingly soluble P fractions in the rhizosphere of white lupin was lower in 0P, 50Pi, and 100Po than in their unplanted control soils in Regosol (Table 5). However, in Andosol, only the NaOH–Po pool decreased, without apparent reduction of the NaOH–Pi fraction in its rhizosphere (Table 6). In the intercropping system, white lupin influenced the maize rhizosphere to alter the concentration of sparingly soluble P pools in 0P, 50Pi, and 100Po applications only in Regosol. Because of the white lupin's effect, maize showed lower concentrations of both NaOH–Pi and NaOH–Po in its rhizosphere in that intercropping system compared to sole cropping (Table 5). The respective degrees of reduction of NaOH–Pi in maize rhizosphere in intercropping systems with the application of 0P, 50Pi, 100Pi, and

Table 4 pH of unplanted control soil and rhizosphere soil of maize and white lupin. Values represent mean of three replicates ±SE (standard error)

P supply	Cropping	Regosol	Andosol
0P	UP	5.42±0.02 a	5.18±0.04 a
	MM	5.43±0.02 a	5.16±0.04 a
	IM	5.24±0.02 ab	5.17±0.09 a
	ML	4.99±0.07 b	4.56±0.02 b
	IL	5.00±0.09 b	4.58±0.07 b
50Pi	UP	5.40±0.03 a	5.19±0.02 a
	MM	5.41±0.02 a	5.20±0.05 a
	IM	5.29±0.02 ab	5.17±0.04 a
	ML	5.00±0.02 b	4.59±0.04 b
	IL	5.00±0.14 b	4.59±0.03 b
100Pi	UP	5.43±0.02 a	5.21±0.03 a
	MM	5.45±0.03 a	5.19±0.08 a
	IM	5.38±0.03 ab	5.17±1.00 a
	ML	5.27±0.03 bc	4.73±0.04 b
	IL	5.22±0.04 c	4.74±0.04 b
100Po	UP	5.52±0.02 a	5.32±0.04 a
	MM	5.53±0.02 a	5.30±0.02 a
	IM	5.37±0.04 b	5.28±0.02 a
	ML	5.14±0.04 c	4.61±0.02 b
	IL	5.15±0.04 c	4.64±0.03 b

0P no P addition, *50Pi* 50 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Pi* 100 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Po* 100 mg P kg^{-1} soil by phytate, *UP* unplanted control, *MM* monocropped maize, *IM* intercropped maize, *ML* monocropped lupin, *IL* intercropped lupin

Different letters in a given column under each P supply denote significant differences ($P \leq 0.05$)

100Po in Regosol were 7.6, 6.7, 1.8, and 9.2 mg P kg^{-1} soil. In the case of NaOH–Po, the respective degrees of reduction were 7.7, 7.8, 2.7 and 15.5 mg P kg^{-1} soil. A tendency to increase the NaOH–Po concentration in maize rhizosphere was noted in Andosol, irrespective of the cropping system (Table 6). The HCl-P fraction was accumulated significantly in white lupin rhizosphere in both soils, where the augmentation appeared to be greater in Andosol (Tables 5 and 6). Maize plants also tended to increase HCl-P fraction in the rhizosphere in Andosol (Table 6). No significant change of residual P fraction was found in maize rhizosphere in either soil (Tables 5 and 6), although a substantial reduction of this pool in lupin rhizosphere in Regosol was noted. The change was not significant (Table 5). The soil total P, which was determined separately, was significantly lower in the rhizospheres of both plants than in unplanted control soil irrespective of the P rate and form,, cropping system, and soil type (Tables 5 and 6).

Discussion

Effects of intercropped white lupin on growth and P accumulation of maize plant

This study demonstrates the effect of white lupin on early stage growth and P uptake of maize plant under different P rates and forms in two contrasting soils. The soil type was a major factor determining the beneficial effects of intercropping because in this study, a complementary effect of mixed cropping was observed only in Regosol (Table 2). This positive effect of intercropping is apparent both in low (0P) and moderate (50Pi) levels of P supply, as well as with organic P application (100Po). In this soil, growth and P accumulation of intercropped maize were higher than those of monocropped maize, suggesting that white lupin increases the availability of P for maize to uptake in mixed culture. Intercropping affected none of these parameters for white lupin, suggesting that P

Table 5 Soil P fractions (mg kg^{-1} soil) of unplanted control soil and rhizosphere soil of maize and white lupin in two cropping systems under the application of different P rates and forms in Regosol. Values represent the mean of three replicates±SE (standard errors)

P supply	Cropping	Resin P	$NaHCO_3$-Pi	$NaHCO_3$-Po	NaOH-Pi	NaOH-Po	HCl-P	Residual P	Total P
0P	UP	15.4±0.7 a	26.4±1.4 a	5.8±0.5 b	38.6±1.6 a	18.5±1.7 a	23.9±2.7 c	125.8±8.4 a	254.4±5.4 a
	MM	10.1±0.7 bc	6.1±0.3 c	2.4±0.3 b	37.5±0.4 a	18.2±1.4 a	26.4±0.9 c	127.1±1.1 a	225.8±3.2 b
	IM	7.8±0.4 c	4.7±0.2 c	2.3±0.3 b	30.9±1.3 b	10.9±1.1 b	27.6±0.8 bc	126.2±2.9 a	210.5±3.4 bc
	ML	11.5±0.7 b	10.8±0.6 b	13.1±1.4 a	18.6±1.1 c	8.1±1.2 b	34.8±1.1 ab	100.8±4.9 a	197.8±6.9 bc
	IL	10.5±0.6 b	9.1±0.9 bc	10.3±0.4 a	17.4±1.4 c	7.2±0.8 b	38.7±0.7 a	101.5±5.8 a	194.8±6.3 c
50Pi	UP	55.6±2.6 a	35.1±1.33 a	5.4±0.3 c	37.9±0.8 a	23.7±1.1 a	19.3±0.9 c	125.5±6.2 a	301.8±5.8 a
	MM	11.2±1.0 c	10.5±0.8 b	2.9±0.3 c	37.1±1.0 a	23.1±1.1 a	22.5±1.3 bc	124.4±2.5 a	232.7±2.4 b
	IM	10.3±0.8 c	10.3±1.2 b	2.6±0.4 c	31.2±2.2 a	15.9±1.2 b	23.7±0.8 bc	125.3±3.1 a	219.2±1.5 b
	ML	35.1±1.3 b	13.4±0.4 b	19.3±1.5 a	20.4±1.9 b	10.8±2.1 b	28.9±0.9 ab	105.4±3.4 a	233.4±2.6 b
	IL	33.5±1.7 b	11.1±1.2 b	14.7±0.5 b	20.9±0.9 b	11.6±0.5 b	34.3±2.1 a	106.3±3.9 a	232.4±2.4 b
100Pi	UP	112.5±3.4 a	37.5±0.6 a	4.1±0.2 b	39.5±0.8 a	26.6±2.7 a	19.4±0.9 b	110.4±8.3 a	350.1±3.6 a
	MM	21.4±0.3 c	19.3±1.1 bc	3.1±0.5 b	40.9±1.3 a	25.8±1.0 a	23.1±0.7 b	113.6±3.4 a	244.2±5.6 c
	IM	20.3±0.7 c	22.0±1.1 b	3.0±1.2 b	37.7±1.5 a	23.9±1.0 a	23.4±0.8 b	112.7±2.3 a	241.1±1.6 c
	ML	42.1±1.5 b	14.1±1.2 d	19.0±2.3 a	30.1±1.5 b	22.3±1.8 a	35.1±1.1 a	105.3±2.6 a	268.0±6.5 b
	IL	42.5±1.5 b	14.7±1.1 cd	20.2±1.2 a	31.1±1.1 b	22.4±2.1 a	31.1±1.2 a	104.3±3.7 a	266.4±4.3 b
100Po	UP	17.8±1.8 a	23.4±0.9 a	11.2±0.4 b	41.2±2.4 a	63.4±1.5 a	19.9±1.3 b	176.3±5.9 a	353.1±5.1 a
	MM	10.3±0.4 b	4.2±0.5 b	5.2±0.4 c	40.5±2.3 a	62.3±1.9 a	22.2±1.1 b	178.6±7.5 a	321.4±5.8 b
	IM	9.4±0.5 b	3.9±0.4 b	5.0±0.2 c	31.9±1.5 a	47.9±2.6 b	27.7±1.4 a	175.4±4.1 a	302.3±5.4 bc
	ML	12.9±1.2 b	5.1±0.5 b	20.4±1.7 a	19.6±1.2 b	37.6±3.5 b	27.5±0.8 a	153.9±2.9 a	277.1±5.9 c
	IL	13.3±0.5 ab	4.9±0.4 b	19.3±0.9 a	18.3±1.5 b	38.6±4.4 b	29.9±1.3 a	155.1±3.9 a	279.3±6.0 c

0P no P addition, *50Pi* 50 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Pi* 100 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Po* 100 mg P kg^{-1} soil by phytate, *UP* unplanted control soil, *MM* monocropped maize, *IM* intercropped maize, *ML* monocropped lupin, *IL* intercropped lupin

Different letters in a given column under each P supply denote significant differences ($P \le 0.05$)

Table 6 Soil P fractions (mg kg^{-1} soil) of unplanted control soil and rhizosphere soil of maize and white lupin in two cropping systems under the application of different P rates and forms in Andosol. Values represent the mean of three replicates±SE (standard errors)

P supply	Cropping	Resin P	$NaHCO_3$-Pi	$NaHCO_3$-Po	NaOH-Pi	NaOH-Po	HCl-P	Residual P	Total P
0P	UP	1.03±0.11 a	21.7±0.4 a	20.8±0.6 a	63.2±0.9 a	129.4±2.8 a	1.9±0.1 d	234.7±14.6 a	472.7±12.7 a
	MM	0.70±0.10 a	5.4±0.7 c	5.7±0.5 c	62.1±1.4 a	137.3±1.4 a	4.1±0.1 c	234.6±5.2 a	449.9±3.2 ab
	IM	0.69±0.10 a	3.7±0.1 c	5.6±0.1 c	61.1±0.5 a	135.4±1.1 a	4.9±0.2 c	236.5±3.9 a	448.1±3.5 ab
	ML	0.87±0.08 a	9.9±1.3 b	15.6±1.6 b	58.0±1.0 a	111.3±2.7 b	9.9±0.1 a	231.3±8.0 a	436.9±7.9 b
	IL	0.87±0.09 a	11.4±0.5 b	12.3±0.6 b	57.9±2.6 a	109.6±3.0 b	8.8±0.6 b	233.3±5.9 a	434.6±6.8 b
50Pi	UP	29.24±2.25 a	32.5±0.5 a	27.6±2.5 a	74.4±3.9 a	130.2±10.9 ab	2.2±0.1 d	227.9±19.6 a	524.1±14.4 a
	MM	13.09±1.35 b	8.2±0.7 c	9.4±0.9 b	77.4±1.6 a	145.2±2.9 a	4.2±0.2 c	229.3±13.9 a	486.9±10.4 b
	IM	10.95±1.12 b	10.3±0.8 c	11.4±0.9 b	73.4±1.3 a	140.7±2.8 ab	5.2±0.3 c	229.9±3.2 a	481.9±1.4 b
	ML	14.14±0.81 b	18.3±1.1 b	17.2±1.1 b	71.2±2.9 a	114.4±4.9 b	9.6±0.7 b	229.1±7.4 a	474.0±5.1 b
	IL	12.25±0.42 b	20.2±1.5 b	14.5±1.3 b	70.3±2.0 a	116.9±1.5 b	11.7±0.5 a	230.1±4.2 a	475.9±5.3 b
100Pi	UP	44.92±2.77 a	43.3±0.8 a	24.7±2.9 a	93.6±2.5 ab	129.4±5.2 bc	2.6±0.1 b	229.5±4.3 a	568.0±1.9 a
	MM	8.69±0.87 c	8.2±0.8 c	7.5±1.2 b	95.6±0.4 a	147.6±3.4 a	3.8±0.4 b	231.5±6.7 a	502.9±7.0 b
	IM	7.81±1.04 c	9.2±0.6 c	9.5±1.1 b	94.6±1.2 a	141.9±0.5 ab	4.9±0.3 b	231.9±7.6 a	499.8±8.7 b
	ML	16.04±0.86 b	17.7±0.8 b	14.7±1.4 b	88.3±1.8 b	116.8±1.4 c	15.9±1.4 a	234.5±7.5 a	504.0±7.9 b
	IL	12.73±1.04 bc	19.9±1.1 b	11.4±1.2 b	90.6±1.4 ab	119.9±4.8 c	14.2±0.6 a	232.5±7.3 a	501.3±9.3 b
100Po	UP	1.23±0.11 a	23.2±0.8 a	19.3±0.5 a	68.6±3.3 a	147.9±2.9 a	2.1±0.1 b	303.9±4.2 a	566.3±9.5 a
	MM	0.73±0.11 ab	5.1±0.6 c	5.2±0.5 c	66.6±1.1 ab	158.6±0.4 a	3.2±0.2 b	305.9±2.4 a	545.5±3.3 b
	IM	0.57±0.07 b	6.2±0.7 c	4.2±0.5 c	65.5±1.8 ab	156.1±2.9 a	4.3±0.5 b	306.1±7.7 a	543.1±11.3 bc
	ML	1.06±0.18 ab	10.9±1.2 b	12.0±1.7 b	61.8±2.1 ab	131.0±3.1 b	12.5±1.1 a	299.9±5.7 a	529.2±4.4 bc
	IL	1.07±0.12 ab	7.8±1.2 bc	8.8±0.7 b	59.4±1.1 b	129.8±2.2 b	15.5±2.2 a	302.1±4.9 a	524.5±5.5 c

0P no P addition, *50Pi* 50 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Pi* 100 mg P kg^{-1} soil by $NaH_2PO_4.2H_2O$, *100Po* 100 mg P kg^{-1} soil by phytate, *UP* unplanted control soil, *MM* monocropped maize, *IM* intercropped maize, *ML* monocropped lupin, *IL* intercropped lupin

Different letters in a given column under each P supply denote significant differences ($P \leq 0.05$)

facilitation occurs from white lupin to maize and white lupin gains no advantage or disadvantage from intercropping. This result also underscores the fact that, in mixed cultures white lupin can obtain adequate P while enhancing P supply for the companion plant.

An interesting finding is that the positive effect of intercropping was evident even under organic P supply as phytate. Our results complement the findings of Li et al. (2003) and Li et al. (2004), who found that chickpea could facilitate P uptake of wheat and maize from the same organic P source; phytate. This study demonstrates the ability of white lupin to facilitate P uptake of companion plant from an organic P source, similar to the facilitation of P to wheat and maize by chickpea. Regarding plant growth, it might be argued that the difference of plant size between maize and white lupin affected the growth and P uptake of maize in both cropping systems through alteration of the competitive relationship between plants because, in this study, maize is somewhat larger than white lupin. This phenomenon suggests that a maize plant grown with a smaller white lupin in a pot filled with 1 kg soil would have ability to explore more resources in soil than a maize plant grown with another maize plant of equal size in the same pot. However, our study did not specifically examine intra-specific competition for soil P between two maize plants and inter-specific competition between maize and white lupin. Consequently, a future experiment that includes a same size pot of single maize plant as a cropping treatment with the other two cropping combinations used in our study must be undertaken to address the potential issue of competitive relation on P acquisition by maize in different cropping systems.

Cluster root formation and rhizosphere acidification by white lupin

Cluster root formation of white lupin (Fig. 1) is consistent with that of a study by Hassan et al. (2012) who reported the formation of many cluster roots by white lupin in a soil culture experiment conducted with the addition of soluble P to a rate of 80 mg-P kg^{-1} soil. Li and Liang (2005), Shane et al. (2003), and Shen et al. (2005) have reported that the formation of cluster roots decreased concomitantly with increasing P concentration in shoots and phloem sap of white lupin at higher P supply. Consequently, the complete suppression of cluster root formation in 100Pi in Regosol occurs because of high shoot P concentration of white lupin. White lupin had a lower shoot P concentration in Andosol than in Regosol, which might have affected differential capacities to form cluster roots in two soils. In the rhizosphere, pH change can be brought about by several processes and rhizosphere acidification caused by carboxylates exudation has been reported from earlier studies. Nuruzzaman et al. (2006) demonstrated the acidification of the rhizosphere of white lupin, which secreted a more substantial amount of citrate than the amounts from other plant species used in that study. Rhizosphere acidification of white lupin was also reported from a study by Cu et al. (2005) in which white lupin reduced the citric-acid-leachable soil P fraction. Therefore, by comparing those previous results with the rhizosphere pH changes in our study, we infer that the observed pH reduction in the rhizosphere of white lupin in this study would have occurred as a result of carboxylate exudation from roots.

P fractions in the rhizosphere

Resin P and $NaHCO_3$–Pi pools are regarded as labile soil P fractions. They are known to be the most available for plant growth (Bowman and Cole 1978; Tiessen and Moir 1993). These two labile pools were reduced in rhizosphere of maize and white lupin in both soils, with the greatest reduction in the maize rhizosphere (Tables 5 and 6). The $NaHCO_3$–Po fraction also declined in the rhizosphere of both plants in Andosol, despite the different rates and forms of P supply. However, in Regosol, this reduction was evident only under organic P application (100Po), although this fraction was augmented in the white lupin rhizosphere. Although the $NaHCO_3$–Po fraction is regarded as labile and available to plants (Tiessen et al. 1992), its availability might be limited by binding to metal oxides in soil (Stewart and Tiessen 1987).

The NaOH extractable Pi and Po, acid extractable (HCl) P and residual P are regarded as less labile pools (Hedley et al. 1982; Tiessen and Moir 1993). Both NaOH–Pi that is P-associated with Al and Fe hydrous oxide (Hedley et al. 1982) and NaOH–Po were decreased significantly in the rhizosphere of white lupin in Regosol, but only NaOH-Po was decreased in Andosol (Tables 5 and 6). Reportedly, P in sparingly soluble fractions can be solubilized by carboxylates (George et al. 2002b; Gerke 1992; Richardson et al. 2001; Wang et al. 2006, 2012). This phenomenon is likely to be relevant to our study for observed rhizosphere pH reduction. Although we did not analyze the organic acid or acid phosphatase activity in rhizosphere soil, the pH decline in white lupin rhizosphere suggests the secretion of organic acids from white lupin and their role in increasing P availability in soil. For organic P to be available for plants, it must be hydrolyzed by phosphatase (Richardson et al. 2001). Cluster root formation can further enhance the P uptake of white lupin attributable to the secretion of extracellular acid phosphatases (Adams and Pate 1992; Neumann et al. 1999, 2000; Ozawa et al. 1995; Wasaki et al. 2003). In our study, we infer that lower concentration of NaOH-Po in lupin rhizosphere compared to unplanted control might have occurred by acid phosphatase secretion, as demonstrated by George et al. (2006), who found a positive correlation between activity of acid phosphatase and reduction of NaOH–Po in the rhizosphere of Tithonia and transgenic clover. Reduction of NaOH–Pi and Po in maize rhizosphere was noted only in mixed culture with white lupin in Regosol, but not in monoculture maize. This fact emphasizes the effect of companion white lupin of changing the concentration of sparingly soluble P pools in maize rhizosphere because, in the same soil, these two particular P pools were decreased in white lupin rhizosphere, irrespective of the cropping system. This decrease also suggests the contribution of organic acids and acid phosphatase secreted by white lupin to increase P availability for maize in intercropping system through rhizosphere-sharing. The tendency of maize to increase the NaOH–Po concentration in its rhizosphere in Andosol might reflect the high microbial activity in Andosol and the build-up of more stable organic P. This tendency is consistent with that described in an earlier report by George et al. (2002a), who found augmentation of NaOH–Po in maize rhizosphere in acidic soil.

Not in maize but in white lupin rhizosphere, a tendency of substantial reduction of stable residual P fraction was apparent, which presumably consists of P that is bound strongly with Al and Fe oxides (Hedley et al. 1982), although the reduction of this pool in lupin rhizosphere is not significant (Table 5). The absolute reduction of residual P in lupin rhizosphere in 0P in Regosol was approximately 25 mg P kg^{-1}. For comparison, under the same P supply and soil, reduction of NaOH-Pi and NaOH-Po were, respectively 20 and 10.4 mg P kg^{-1}. However, considering the concentration of a particular P pool in unplanted control soil under the P supply and soil conditions described above, residual P recorded the highest concentration of 125.8 mg P kg^{-1} whereas the concentrations of NaOH-Pi and NaOH-Po were, respectively 38.6 and 18.5 mg P kg^{-1} (Table 5). Consequently, this high base value and low proportion of the depletion of residual P compared to other P pools, probably coupled with low number of replication (n=3) would have caused this change insignificant in our study. Acid phosphatase by white lupin might have influenced this reduction in lupin rhizosphere.

Enhanced growth and P benefits for maize from intercropping with white lupin differ among contrasting soils and different P rates and forms

Our findings of positive effect of intercropped white lupin on maize in Regosol, an acidic soil show agreement with results reported by Cu et al. (2005) in alkaline soil. However, our study has produced the novel finding that maize can enhance growth and P uptake from intercropped white lupin only in Regosol: not in Andosol. The differences observed in intercropping effect on maize in two contrasting soils are explainable mainly by three factors: (1) difference in P availability between two soils as affected by their P adsorption capacities; (2) contrasting root growth behavior of maize and root intermingling of maize with white lupin between two soils; and (3) differential capacity of white lupin to increase P availability for intercropped maize to uptake between two soils. In the intercropping system, roots of maize and white lupin can mix together. For that reason, strong root intermingling complements companion maize plant to uptake more P, which is made available by white lupin. Strong root intermingling is evident from the rhizosphere pH reduction of intercropped maize in Regosol. Full root intermingling between two plants grown in a small pot filled with 1 kg soil can be expected in any soil type if crop growth is normal. Both high bulk density and low water holding capacity of Regosol compared to Andosol suggest that Regosol is a heavier and more compacted soil than Andosol is (Table 1). Although root elongation of maize could be easier under these conditions, we observed poorer root growth in Andosol (Table 3). Therefore, this contrasting root growth behavior in two soils highlights the fact that, the Andosol itself has limited the root growth and development of maize. The root architecture of plants can undergo several changes in response to P deficiency. Effects of soil P availability for the root growth of maize have been demonstrated previously. Hajabbasi and Schumacher (1994) have reported a significant negative effect of soil P availability on the rate of appearance of axile roots of maize. Mollier and Pellerin (1999) showed reduction in both lateral root elongation and emergence of new axile roots of maize under low P availability. This phenomenon is clear from comparison of P availability of two soils used in present study, where in Andosol, resin P, which is regarded as readily available fraction for plants is extremely low (Table 1). For comparison, resin-P concentrations in unfertilized crop and pasture soils are in the range of 20–40 mg P kg^{-1} dry soil (Hedley et al. 1982). Ten times higher phosphorus adsorption capacity of Andosol than Regosol would explain the high fixation of applied P in less labile pools. Many Andosols are rich in allophanes, containing active aluminum and show a high retention of P, which makes the utilization of applied fertilizer P particularly low (Lukito et al. 1998) because accumulated P exists mainly in non-labile forms (Hirata et al. 1999). This phenomenon is expected to have led to the poorer root growth of maize in Andosol.

The concentrations of three less labile pools, NaOH–Pi, NaOH–Po, and residual P were lower in lupin rhizosphere in Regosol (Table 5) compared to Andosol, where the notable reduction was occurred only from one less labile pool, NaOH–Po (Table 6). P pools can decrease by root uptake but can also increase or decrease by shifts of P from one pool to another, through changing rhizosphere chemistry, induced by root exudates. Microbial processes that are effective in directly solubilizing and mineralizing P from organic P can also change the concentrations of P pools in soil (Richardson et al. 2009). Most probably, exudation of P-mobilizing compounds of white lupin affects this reduction and part of the P declined from above pools would have transferred to labile P pools as an increment of $NaHCO_3$-Po pool in lupin rhizosphere in Regosol. If this happens, it suggests that the ability of white lupin to increase P availability is higher in Regosol than in Andosol, which constitutes another contributing factor that complements intercropped maize to uptake more P in Regosol, creating the observed difference between two soil types used in the study.

Although positive effects of intercropped white lupin for main crop plant are achieved in a soil, the application of high doses of soluble P to that particular soil can eliminate this complementary effect because the ability of white lupin to increase P availability apparently declines with increasing soluble P supply. Moreover, the main crop plant would be able to uptake the required amount of P under high soluble P supply without the supportive effect of white lupin. This phenomenon is readily apparent with the absence of positive effects for intercropped maize under 100Pi in Regosol, although, in the same soil, enhanced growth and P acquisition by intercropped maize with white lupin were found for 0P, 50Pi supply and with the addition of P as an organic form: phytate.

Conclusions

Results of this study demonstrate that interspecific P facilitation in the intercropping of maize and white lupin is unidirectional, occurring from white lupin to maize, where white lupin increases the availability of P and companion maize uptake part of those, giving no benefit to white lupin. Enhanced growth and P accumulation by maize with the effect of intercropped white lupin can be dependent on the soil type where they are grown and rates and forms of P applied to particular soil. Findings also highlight the ability of white lupin to improve growth and P acquisition of maize in mixed culture in Regosol without detrimentally affecting its own growth or P accumulation. Nevertheless, white lupin is not the ideal P-efficient plant to improve companion P-inefficient plant species in some soils. Our results open the door to additional detailed exploration of this model cropping system in actual field conditions in Regosol, where crops are grown to maturity and where translocation of P to the grain occurs, to ascertain whether white lupin can benefit companion maize plant in the latter stage of growth, as observed in the early stage of growth in our study. Before a definitive conclusion can be made, more field studies conducted with widely various soil types are warranted to confirm the applicability of white lupin as a P-efficient plant to improve P-inefficient plants in mixed cropping systems.

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