

Genotypic Variation in P Utilization of Soybean(*Glycine max* L.) Grown in Various Insoluble P Sources

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Abstract The information on soybean genotypes originating from different pH soil utilized insoluble P sources was various. Three soybean genotypes representing typical native species were collected from south, western and northeast of China. Plant biomass, P accumulation, organic acid excretion, pH and PUE were analysed. Three genotypes all favoured in Al-P, however, Suinong 10 and Nibadou total biomass in Al-P was 8.5% and 9.4% less than Na-P, respectively, then 5.3% and 11.8%, 6.4% and 42.2% more than Ca-P and Fe-P, respectively. For Xin soybean 1 total biomass was 1.4%, 20.8% and 40.8% more than Na-P, Fe-P and Ca-P, respectively. P concentration ranged from 1.37 to 2.47 mg/g, 1.39 to 3.04 mg/g and 3.20 to 4.73 mg/g in shoots, roots and nodules, respectively. The maximum total P concentration was Al-P, Fe-P and Fe-P in Suinong10, Xin soybean 1 and Nibadou, respectively. Irrespective of genotype, the amount of organic acid exuded followed the order L(-)-malic>Oxalic>L(+)-lactic>Malonic>Succinic>Citric. However, for Suinong 10 the order was Oxalic>L(+)-lactic>L(-)-malic, for Xin soybean 1 the order was L(+)-lactic>L(-)-malic>Malonic>Oxalic>Citric and for Nibadou the order was L(-)-malic>Oxalic>L(+)-lactic>Citric>Malonic. Solution pH ranged from 5.48 to 6.52. PUE (phosphorus use efficiency) in Xin soybean 1 was more than Suinong 10 and Nibadou, irrespective of P source. The maximum PUE was Al-P, Al-P and Na-P in Suinong 10, Xin soybean 1 and Nibadou. The results suggest that soybean genotypes differ in growth response and P uptake from insoluble P sources and genotypic variation in P acquisition was related to root exudation and pH.

Key words Genotypic variation; P utilization; Insoluble P; Soybean

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大豆利用难溶磷源基因型差异

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摘要 来源于不同 pH 土壤上的各种大豆基因型利用难溶性磷源有明显差异。采用分别代表着南

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方、西北和东北本地品种三种基因型大豆,研究它们生物量、磷素积累、分泌的有机酸、根际 pH 和磷素利用效率(PUE)的差异。三种基因型大豆都比较偏爱 Al-P,然而,绥农 10 号和泥巴豆在利用 Al-P 时,生物量比 Na-P 分别少 8.5%和 9.4%,比 Ca-P 和 Fe-P 分别多 5.3%和 11.8%,6.4%和 42.2%。新大豆 1 号利用 Al-P 时生物量比 Na-P、Fe-P 和 Ca-P 分别多 1.4%、20.8%和 40.8%。地上部、根系和根瘤含磷量变化范围分别在 1.37~2.47 mg/g、1.39~3.04 mg/g 和 3.20~4.73 mg/g。绥农 10 号、新大豆 1 号和泥巴豆最大总磷含量分别出现在 Al-P、Fe-P 和 Fe-P 条件下。泥巴豆根系分泌的有机酸表现出下面的顺序:草酸>乳酸>苹果酸;新大豆 1 号表现为:乳酸>苹果酸>丙二酸>草酸>柠檬酸;泥巴豆表现为:苹果酸>草酸>乳酸>柠檬酸>丙二酸。无论供给哪种形态磷源,根际 pH 变化范围为 5.48~6.52。新大豆 1 号磷素利用效率比绥农 10 号和泥巴豆高,绥农 10 号、新大豆 1 号和泥巴豆最大磷素利用效率分别出现在 Al-P、Al-P 和 Na-P 源条件下。这些结果表明,不同基因型大豆生长和磷吸收对各种难溶磷源的反应不同,各基因型磷素利用差异与根系分泌物和根际 pH 有关。

关键词 基因型变化;磷素利用;难溶磷源;大豆

Phosphorus (P) was one of the least available mineral nutrients to plants in many agricultural environments [1]. However, many metabolic processes, such as energy transfer, photosynthesis, respiration and signal transduction, etc., need P participation. Due to least available, least mobile, P availability determines plant growth. Otherwise, temperature, moist and geography factors affected P-type reserved in different terrestrial ecology. Plant species differ in the efficiency with which they acquire P. P acquisition efficiency relates to the different extent to which plants were able to mobilize from poorly soluble sources or to take up the soluble P available in the soil solution [2]. The P uptake was a multigenic with involvement of additive, dominance and epistatic effects [3]. Plant modifies many mechanisms to increase P acquisition, uptake, absorption efficiency, such as root morphology [4], proteoid root [5~7], organic acid exudation [8,9] etc. It is known that organic acid exudation by rape roots was an effective strategy to increase P uptake from applied rock phosphate [10]. Of the acids secreted, citric and oxalic acids were the most efficient at solubilizing P [11].

Low P was the major constraints to soybean growth on large reserves total P agricultural soil in China. Especially, the major P source from south to north of China was different, thus identify genotypic

variation in P utilization of soybean growth in various insoluble P source was very important for screening higher P efficiency genotype, furthermore, leading to decrease exogenous P fertilization application for plant growth, and paving way to economically and ecologically sustainable cropping systems [1].

1 Materials and methods

1.1 Growth conditions

A sand with extremely low in both available P (<1 mg/kg) and total P was used. Three kg of sand was weighed into each of plastic bags. Basal nutrients were added in stock solution onto the surface of the sand. Composition of the solution was as the follows (in mol/L): K_2SO_4 , 133; $CaCl_2 \cdot 2H_2O$, 167; $MgSO_4 \cdot 7H_2O$, 43; H_3BO_3 , 0.67; $MnSO_4 \cdot H_2O$, 6.7; $ZnSO_4 \cdot 7H_2O$, 10; $CuSO_4 \cdot 5H_2O$, 2; $CoSO_4 \cdot 7H_2O$, 0.33; $Na_2MoO_4 \cdot 2H_2O$, 0.17. Four P sources: $FePO_4$, $AlPO_4$, Ca-P (hydroxyapatite) and NaH_2PO_4 (as P-sufficient control). All P sources were added at a rate of 25 mg P/kg soil. After re-dried, the P chemicals were added in powder to the designated treatment sands. The sand was then mixed thoroughly with basal nutrients and P by shaking the sand in a jar. Four replicates. All the pots were watered to field capacity. Sand moisture was maintained by

watering the pots to the designated weight (from bottom). Distilled water was used throughout the experiment.

1.2 Plants materials

Three typical native genotypes, originating from northeast, western and south of China, were Suinong 10, Xin soybean 1 and Nibadou, respectively. Ten uniform-sized germinated seeds were sown to each pot at 1 cm depth. 1 mL of rhizobium suspension was added onto each seed before it was covered by sand. When the first pair of leaves was fully expanded, plants were thinned to four per pot. To minimize the effect of seed P reserve, cotyledons of all the plants were removed after first pair of true leaves were fully expanded. Plants harvested 90d after sowing (Plant growth response to P was obvious).

Plant separated into shoots, roots and nodules. Biomass of all parts was measured. Plant tissues were oven-dried at 80 C to constant weight for P and N analysis. The total P digested by H₂SO₄ and H₂O₂ was analysed with ultraviolet spectrometer (UV2500 Japan) and N concentration in plant tissues was analysed by auto-titration after H₂SO₄ and H₂O₂ digestion^[12]. Organic acid exuded was collected by leaching through the pots with distilled water, using HPLC to analyse total amount of organic acid and species. The pH of rhizosphere solution was measured by pH measurement.

2 Results

2.1 Plant growth

The effects of P sources and genotypes on biomass of shoots, and nodules were significant (Table 1). For Suinong10, shoot biomass ranged 2.65–3.68 g per plant in an order: Na-P > Al-P > Ca-P > Fe-P; root biomass ranged 0.67–0.74 g per plant in an order: Fe-P > Na-P > Al-P > Ca-P and nodule biomass ranged 0.17~0.20 g per plant in an order: Ca-P > Al-P > Na-P > Fe-P. For Xin soybean 1, shoot biomass ranged 3.68–5.37 g per plant in an order: Na-P > Al-P > Fe-P > Ca-P; root biomass ranged 1.16~1.56 g per plant in an order: Al-P

> Na-P > Fe-P > Ca-P and nodule biomass ranged 0.19~0.28 g per plant in an order: Al-P > Na-P > Fe-P > Ca-P. For Nibadou, shoot biomass ranged 2.44~3.88 g per plant in an order: Na-P > Al-P > Ca-P > Fe-P; root biomass ranged 0.67~1.06 g per plant in an order: Na-P > Al-P > Ca-P > Fe-P and nodule biomass ranged 0.16~0.20 g per plant in an order: Na-P > Al-P > Ca-P > Fe-P. In comparison, total biomass was Xin soybean 1 > Nibadou > Suinong 10, irrespective P sources. The shoot biomass was much more affected by P source and genotype than root and nodule. Thus, Na-P was the optimal P source for three plants biomass. Otherwise, Suinong 10 and Nibadou favoured in Al-P, though its total biomass was 8.5% and 9.4% less than Na-P respectively, 5.3% and 11.8%, 6.4% and 42.2% more than Ca-P and Fe-P respectively. For Xin soybean 1 the optimal P were similar to that for Suinong 10. Total biomass of Xin soybean was 1.4%, 20.8% and 40.8% more than Na-P, Fe-P and Ca-P respectively.

Table 1 Dry weights of shoots, roots and nodules of different P sources-fed of Suinong 10, Xin soybean 1 and Nibadou at 40 days after sowing

Genotype	P sources	Tissues biomass (g/plant)		
		Shoots	Roots	Nodules
Suinong 10	Ca-P	2.88	0.67	0.20
	Al-P	3.11	0.69	0.19
	Fe-P	2.65	0.74	0.17
	Na-P	3.44	0.73	0.18
Xin soybean 1	Ca-P	3.68	1.16	0.19
	Al-P	5.24	1.56	0.28
	Fe-P	4.33	1.34	0.20
	Na-P	5.37	1.36	0.26
Nibadou	Ca-P	3.21	0.96	0.19
	Al-P	3.46	1.01	0.18
	Fe-P	2.44	0.67	0.16
	Na-P	3.88	1.06	0.20
LSD (P=0.05) for any two means significance level		0.58	0.28	0.05
Genotype		* * *	* * *	* * *
P source		* *	*	*
Genotype×P source		* *	*	*

2.2 Phosphorus and nitrogen accumulation in plants

Tissues phosphorus content was significantly affected by P source and genotype, but the interaction between P source and genotype on nodule P content was significant. In comparison, the effect of genotype on phosphorus content in shoots and nodules was more than which in roots, however, phos-

phorus source had more effect on phosphorus content in shoots and roots than in nodules (Table 2). P content ranged from 1.37 to 2.47 mg/g, 1.39 to 3.04 mg/g and 3.20 to 4.73 mg/g in shoots, roots and nodules, respectively. Total P content ranged from 7.59 to 8.55 mg/g, 6.38 to 8.81 mg/g and 7.61 to 8.68 mg/g in Suinong 10, Xin soybean 1 and Nibadou, respectively. The maximum total P

content was Al-P, Fe-P and Fe-P in Suinong10, Xin soybean1 and Nibadou, respectively. In general, N content in plant tissues was in an order: nodules > shoots > roots. However, genotype significantly affected N content in shoots, and the interaction between genotype and P source also significantly affected that in nodules (Table 2).

Table 2 N and P content in plant tissues of different P sources—fed of Suinong 10, Xin soybean 1 and Nibadou at 40 days after sowing

Genotype	P sources	N content (mg/g)			P content (mg/g)		
		Shoots	Roots	Nodules	Shoots	Roots	Nodules
Suinong 10	Ca-P	30.37	12.65	51.37	2.13	1.47	3.99
	Al-P	31.60	12.99	52.06	2.47	1.35	4.73
	Fe-P	32.54	14.38	46.63	1.77	2.71	3.70
	Na-P	29.50	14.81	50.58	2.44	1.82	3.88
Xin soybean 1	Ca-P	21.85	12.89	49.79	1.52	1.66	3.20
	Al-P	9.64	14.85	50.02	1.37	1.73	3.32
	Fe-P	24.31	14.98	48.66	1.58	3.04	4.19
	Na-P	23.72	14.74	53.61	1.87	1.75	4.30
Nibadou	Ca-P	28.19	14.16	52.96	2.22	1.51	4.51
	Al-P	28.42	13.85	51.55	2.00	1.49	4.12
	Fe-P	31.78	15.24	51.26	1.75	2.73	4.20
	Na-P	26.54	13.49	49.46	2.43	1.39	4.72
LSD (P=0.05)		5.23	3.16	4.06	0.45	0.45	0.51
for any two means Significance level							
Genotype		* * *	ns	ns	* * *	*	* * *
P source		ns	ns	ns	* *	* * *	*
Genotype P source		ns	ns	*	ns	ns	* * *

2.3 Organic acid exuded and rhizosphere pH

The response of total organic acid exuded by root in Suinong 10, Xin soybean 1 and Nibadou to

different P source was shown in Fig. 1. For Suinong 10 followed the order Na-P > Al-P > Fe-P > Ca

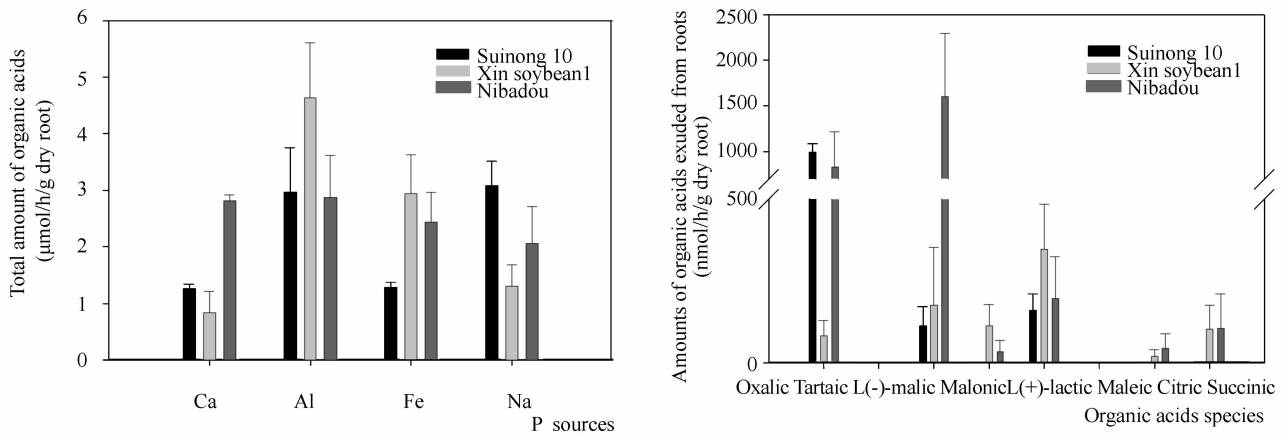


Fig. 1 The amount and species of organic acid of different P sources—fed of Suinong 10, Xin soybean 1 and Nibadou at 40 days after sowing. The bar presents SE at P < 0.05

-P, for Xin soybean 1 the order was Al-P>Fe-P>Na-P>Ca-P and for Nibadou the order was Al-P>Ca-P>Fe-P>Na-P. However, the organic acid species exuded by root was different (Fig. 1). Organic acid exuded followed the order L(-)-malic>Oxalic>L(+)-lactic>Malonic>Succinic>Citric, respectively genotypes. However, for Suinong 10 the order was Oxalic>L(+)-lactic>L(-)-malic, for Xin soybean 1 the order was L(+)-lactic>L(-)-malic>Malonic>Oxalic>Citric and for Nibadou the order was L(-)-malic>Oxalic>L(+)-lactic>Citric>Malonic. Solution pH ranged from 5.48 to 6.52 (Fig. 2). For Suinong 10 followed the order Na-P>Al-P>Ca-P>Fe-P, but for Xin soybean 1 the order was similar to that for Suinong 10. For Nibadou the order was Na-P>Ca-P>Al-P>Fe-P. The relationship between rhizosphere

pH and N,P contents was significant in Al-P. The relationship was similar to that for P content in Ca-P, however, no significant relationship was found in Na-P and Fe-P (Fig. 3).

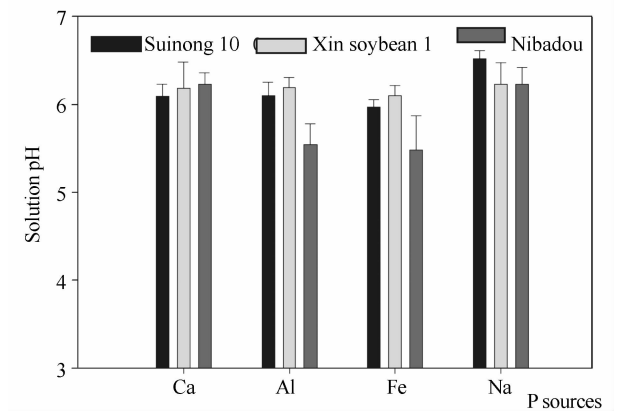


Fig. 2 The rhizosphere pH of different P sources-fed of Suinong 10, Xin soybean 1 and Nibadou at 40 days after sowing. The bar presents SE at $P < 0.05$

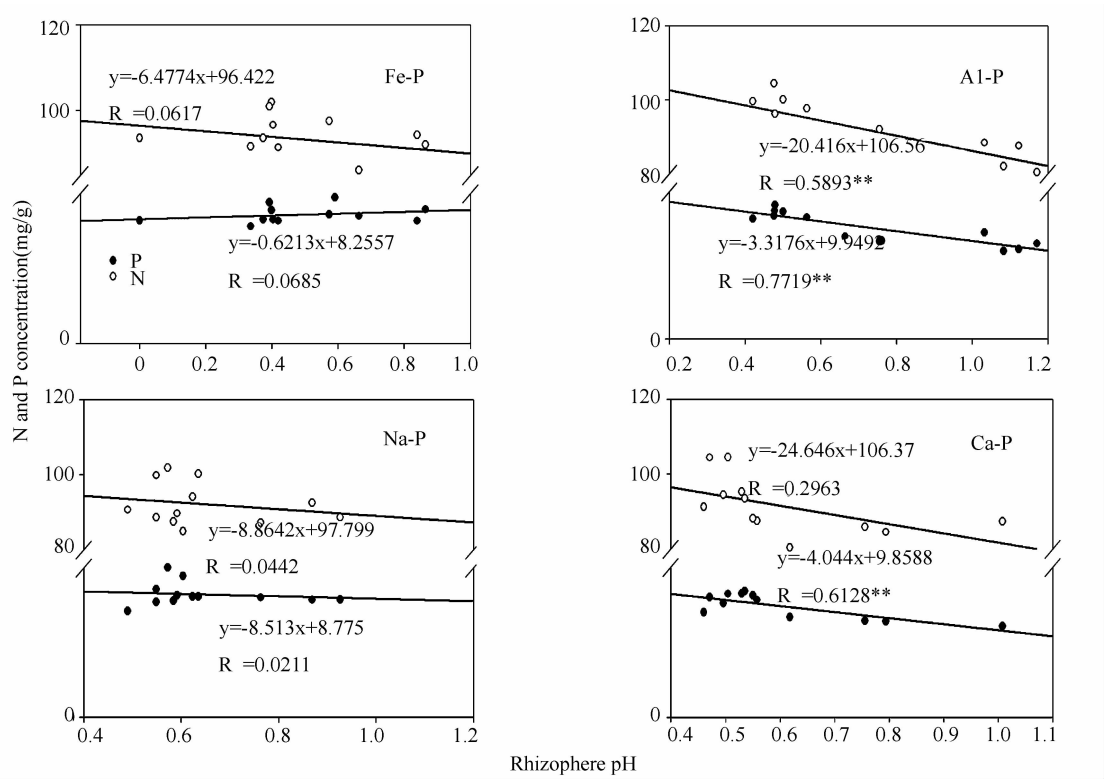


Fig. 3 The relationship between N,P content and rhizosphere pH of different P sources-fed of Suinong 10, Xin soybean 1 and Nibadou at 40 days after sowing

2.4 Phosphorus utility efficiency

In order to assess the efficiency in utilization of P, the phosphorus utilization efficiency (PUE) was computed. The data in Fig. 1 show that PUE in

Xin soybean 1 was more than Suinong 10 and Nibadou, irrespective of P source. The maximum PUE was Al-P, Al-P and Na-P in Suinong 10, Xin soybean 1 and Nibadou, respectively (Fig. 4).

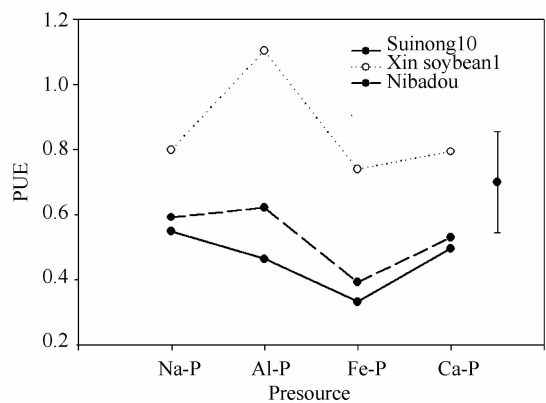


Fig. 4 The PUE of different P sources-fed of Suinong 10,Xin soybean 1 and Nibadou at 40 days after sowing. The bar presents LSD at P < 0.05

3 Discussion

Soybean genotypes differ in growth response and P uptake from insoluble P sources. It can be shown in this experiment (Table 1,2). Variations among plant species in ability to absorb fertilizer phosphate have received ample attention in recent years. It was evident that rock phosphates dissolve more readily in acid than in neutral and alkaline soils for plant to use [13], and acid-forming fertilizers such as Al-phosphates dissolve more readily in alkaline and neutral soil than in acid soils, and enhance plant P availability [14,15,16]. In this sand culture conditions, there were extensive body water in contact with added phosphate fertilizer allows the total quantity of dissolved phosphate to be enough to meet the phosphate requirement of the soybean growth. Thus, the optimal insoluble P source was Al-P for soybean growth, except Na-P. Especially for Xin soybean 1 originated from alkaline conditions, the biomass in Al-P was more than Na-P. It suggested that insoluble Al-P was easier dissolved in the neutral condition. However, insoluble Ca-P did not increase the total biomass for Nibadou, it maybe due to the low level of proton and to Ca common ion affected insoluble Ca-P solubility [16,17]. Otherwise, the P accumulation and PUE were different among three genotypes and four P

sources. Though the relationship between PUE and P accumulation was not significant, on average, higher PUE plant decreased P accumulation in tissues. All of these shown that P source, genotype, the interaction of P source and genotype affected significantly biomass and P content, except that no significant effects of the interaction of P source and genotype on P content (Table 1,2).

Genotypic variation in P acquisition was related to root exudation and pH. In general, legumes were more efficient than other plants in producing and excreting organic acids to enhance Pi solubilization under Pi deficiency [5]. Organic acids exudation included many species,our results showed that L (−)-malic was the most, approximately was 2-fold Oxalic,3-fold L (+)-lactic,5-fold succinic and 10-fold Malonic, respectively. This was same as that in rape [5] and chickpea [18]. Acid production by legume plants varies between species [19~22]. A large variation in response to P source supply existed between the species,especially for Xin soybean 1 total amount of organic acids supplied with Al-P was the most,for Suinong 10 and Nibadou total amount of organic acid was similar to that for Xin soybean 1. The relationship between PUE and amount of organic acid or pH was not significant, but the relationship between N,P content and rhizosphere pH reached significantly with Al-P supply (Fig. 3),thus soybean had a higher PUE with Al-P source. These suggested that organic acid excretion is an efficient strategy for sparingly-soluble P source [23].

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