RESEARCH ARTICLE

Effect of Phosphorus Fertilization to P Uptake and Dry Matter Accumulation in Soybean with Different P Efficiencies

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Abstract

Phosphorus (P) is an essential element for plant growth and yield. Improving phosphorus use efficiency of crops could potentially reduce the application of chemical fertilizer and alleviate environmental damage. Soybean (Glycine max (L.) Merr.) is sensitive to phosphorus (P) in the whole life history. Soybean cultivars with different P efficiencies were used to study P uptake and dry matter accumulation under different P levels. Under low P conditions, the P contents of leaf in high P efficiency cultivars were greater than those in low P efficiency cultivars at the branching stage. The P accumulation in stems of high P efficiency cultivars and in leaves of low P efficiency cultivars increased with increasing P concentration at the branching stage. At the late podding stage, the P accumulation of seeds in high and low P efficiency cultivars were 22.5 and 26.0%, respectively; and at the mature stage were 69.8 and 74.2%, respectively. In average, the P accumulation in whole plants and each organ was improved by 24.4% in high P efficiency cultivars compared to low P efficiency cultivars. The biomass between high and low P efficiency cultivars were the same under extended P condition, while a significant difference was observed at late pod filling stage. At the pod setting stage, the biomass of high P efficiency cultivars were significant greater (17.4%) than those of low P efficiency cultivars under high P condition. Meanwhile, under optimum growth conditions, there was little difference of biomass between the two types of cultivars, however, the P agronomic efficiency and P harvest index were significant higher in high P efficiency cultivars than those in low P efficiency cultivars.

Key words: Glycine max (L.) Merr., soybean, phosphorus use efficiency

INTRODUCTION

A deficiency of available phosphorus (P) in soil is one of the most important factors that limit soybean development and yield (Wissuwa 2003). To maintain crop production, more and more commercial P fertilizers are applied, generating enormous costs, consuming limited phosphate ore resources and destroying the ecological balance and resulting in severe environmental damage (Quan and Yan 2002; Zhang et al. 2005). The total P content is high in soils, but the available P content is low; which leads to genetic P deficiency (Epstein et al. 1983). It is estimated that, the production of germplasm with efficient uptake, transportation and utilization of P...
becomes a topic of global focus (Zhang et al. 2005).

Soybean, a major grain and forage crop, is influenced by genetic P deficiency. In recent years, it is focused on the screening, identification, inheritance, and biochemical mechanisms underlying P use efficiency (Tong et al. 2001; Liang et al. 2006; Liu et al. 2008). So far, most of our knowledge on P use efficiency has come from the biological properties of root system and the yield of soybean; some questions, such as the rates of P uptake, distribution and dry matter accumulation in different P efficiencies genotypes at various growth stages have been rarely reported. It is shown that the plant biomass, soybean yield and P uptake enhanced, with the application of P (Goswami et al. 1999; Andraski et al. 2003). Nutrient accumulation was closely related with the yield of soybean (Zha et al. 2006). The biomass of soybean was also the foundation for the grain yield. Thus, the pattern of distribution of biomass was a biological characteristic that depended on genotype and affected individual development and population yield (Huang et al. 2009; Ao et al. 2013). Cai et al. (2004) indicated that the dry matter content was greatly influenced by the application of P. Each soybean organ responded to the P application, but excess P suppressed soybean growth (Cai et al. 2004). Studies on winter wheat, sorghum, and corn of high P use efficiency cultivars showed that the re-transport and re-use of P were higher, and under low P condition, the plants mentioned above could grow and develop normally, produce much higher grain yield (George 1985; Gstdiner and Christensen 1990). There were significant differences in the abilities to adapt low P condition among soybean cultivars (Huang et al. 2009). It was shown that under low P conditions, the P content of a single plant was much higher in high P use efficiency soybean cultivars, the higher biomass and grain yields of those were the important characteristics under P stress. 

Soybean is a P dependent crop, and application of proper P concentrations coordinated production, improved physiological characteristics, and enhanced nutrient uptake (Yan et al. 1995). P is an essential element for photosynthesis in soybean. The yield of soybean mainly depended on the accumulation, distribution, transportation, and conversion of photosynthesis products (Ao et al. 2009). Therefore, studies of the properties of P uptake, dry matter accumulation in soybean under different P levels and regulation of P in cultivars with various levels of P use efficiency will provide a sound theoretical basis for improvement of soybean yield.

RESULTS

The accumulation and distribution of P in soybean with different P efficiencies at different growth stages

<table>
<thead>
<tr>
<th>Type of P efficiency</th>
<th>P content (%)</th>
<th>P accumulation (mg/plant)</th>
<th>P distribution rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Stem</td>
<td>Leaf</td>
<td>Stem</td>
</tr>
<tr>
<td>High P efficiency</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low P</td>
<td>0.32±0.02 a</td>
<td>0.42±0.025 a</td>
<td>9.90±0.34 a</td>
</tr>
<tr>
<td>Medium P</td>
<td>0.31±0.071 a</td>
<td>0.36±0.074 a</td>
<td>12.55±2.25 a</td>
</tr>
<tr>
<td>High P</td>
<td>0.28±0.012 b</td>
<td>0.37±0.036 a</td>
<td>12.99±1.38 a</td>
</tr>
<tr>
<td>Low P efficiency</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low P</td>
<td>0.23±0.046 b</td>
<td>0.41±0.042 a</td>
<td>10.72±0.55 a</td>
</tr>
<tr>
<td>Medium P</td>
<td>0.25±0.089 b</td>
<td>0.41±0.042 a</td>
<td>10.72±0.55 a</td>
</tr>
<tr>
<td>High P</td>
<td>0.23±0.081 b</td>
<td>0.38±0.011 a</td>
<td>10.83±3.79 a</td>
</tr>
</tbody>
</table>

Values are mean±SD. The different letters indicate significance of treatments at the 5% level. The same as below.
with the increased P concentration (Table 2). The degree of increment on P accumulation in leaves was significantly different between these low and high P efficiency cultivars. In the high P efficiency cultivars, P accumulation in leaves increased by 64.6%, of which increased by 109.6% in the low P efficiency cultivars. In the high P efficiency cultivars, the P distribution rate of stems was the highest under low P concentrations, and it was the lowest under high P concentrations, vice versa in leaves. In the low P efficiency cultivars, the P distribution rate of stems was quite high under optimum P concentration, which of leaves was the highest under low P concentrations. Compared to the two types of cultivars, P accumulation of stems in the high P efficiency cultivars was significant, with a rate of 47.1% higher than that of the low P efficiency cultivars ($P<0.05$). Under medium and high P conditions, the P distribution rate of high P efficiency cultivars was lower than that of the low P efficiency cultivars in stems ($P<0.05$). Although, this value was much higher in leaves of high P efficiency cultivars than that of low P efficiency cultivars ($P<0.05$).

**Podding stage** With the increased P concentration, the P content changed slightly in stems and leaves of the high P efficiency cultivars (Table 3). However, the P content tended to rise in stems and leaves in the low P efficiency cultivars. Compared to the two types of cultivars, the P content and P accumulation of stems and leaves in high P efficiency cultivars was relatively high ($P<0.05$). In low P efficiency cultivars, there was a significantly increased P content of stems under medium and high P treatments than that under low P treatment ($P<0.01$). In high P efficiency genotypes, the P accumulation of leaves was enhanced by 32.4% ($P_{medium}<0.01$) under medium P treatments, and was enhanced by 25.5% ($P_{high}<0.01$) under high P treatments than that under low P treatment. In the low P efficiency cultivars, the P accumulation of stems and leaves was improved under moderate and high P treatments than that under low P treatment, with an average increment of 56.6% ($P_{medium}<0.01$) in stems and 63.3% ($P_{high}<0.01$) in leaves, respectively. Under low P conditions, the P content and P accumulation of stems and leaves in high P efficiency cultivars were much higher than those in low P efficiency cultivars ($P<0.05$). Under optimum P concentrations, the P distribution in leaves of the high efficiency cultivars was significantly higher than that of the low P efficiency cultivars ($P<0.05$), vice versa in stems ($P<0.05$).

**Late podding stage** At the late podding stage, P concentration had slight effect on P content of various soybean organs with high P efficiency (Table 4). With the improved P concentration, the P contents of stems, leaves, pod shells, and seeds in soybean with low efficiency increased. There was a significant difference in P content under high P treatment. With the improved P concentration, P accumulation of various organs increased in both types of soybean, although the degrees were significantly different.

### Table 2 Distribution of P in soybean cultivars with various P efficiencies cultivars at bloom stage

<table>
<thead>
<tr>
<th>Type</th>
<th>P level</th>
<th>P content (%)</th>
<th>P accumulation (mg)</th>
<th>P distribution rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Stem</td>
<td>Leaf</td>
<td>Stem</td>
</tr>
<tr>
<td>High P efficiency</td>
<td>Low P</td>
<td>0.33±0.021 a</td>
<td>0.46±0.036 bc</td>
<td>39.18±3.40 b</td>
</tr>
<tr>
<td></td>
<td>Medium P</td>
<td>0.31±0.028 ab</td>
<td>0.46±0.032 bc</td>
<td>42.52±3.33 b</td>
</tr>
<tr>
<td></td>
<td>High P</td>
<td>0.31±0.013 ab</td>
<td>0.51±0.050 a</td>
<td>40.79±5.56 b</td>
</tr>
<tr>
<td>Low P efficiency</td>
<td>Low P</td>
<td>0.27±0.022 b</td>
<td>0.35±0.021 c</td>
<td>36.64±4.32 c</td>
</tr>
<tr>
<td></td>
<td>Medium P</td>
<td>0.30±0.028 ab</td>
<td>0.43±0.056 c</td>
<td>40.31±5.02 b</td>
</tr>
<tr>
<td></td>
<td>High P</td>
<td>0.34±0.069 a</td>
<td>0.46±0.017 bc</td>
<td>59.00±2.25 a</td>
</tr>
</tbody>
</table>

### Table 3 Distribution of P in soybean cultivars with various P efficiencies at podding stage

<table>
<thead>
<tr>
<th>Type</th>
<th>P level</th>
<th>P content (%)</th>
<th>P accumulation (mg)</th>
<th>P distribution rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Stem</td>
<td>Leaf</td>
<td>Stem</td>
</tr>
<tr>
<td>High P efficiency</td>
<td>Low P</td>
<td>0.29±0.054 a</td>
<td>0.49±0.074 a</td>
<td>54.39±7.03 b</td>
</tr>
<tr>
<td></td>
<td>Medium P</td>
<td>0.32±0.038 a</td>
<td>0.49±0.032 a</td>
<td>62.27±10.28 ab</td>
</tr>
<tr>
<td></td>
<td>High P</td>
<td>0.31±0.022 a</td>
<td>0.46±0.01 ab</td>
<td>63.14±3.72 ab</td>
</tr>
<tr>
<td>Low P efficiency</td>
<td>Low P</td>
<td>0.24±0.010 b</td>
<td>0.42±0.060 b</td>
<td>40.63±1.31 c</td>
</tr>
<tr>
<td></td>
<td>Medium P</td>
<td>0.29±0.035 a</td>
<td>0.44±0.051 b</td>
<td>64.77±9.62 a</td>
</tr>
<tr>
<td></td>
<td>High P</td>
<td>0.29±0.027 a</td>
<td>0.47±0.039 ab</td>
<td>62.45±6.89 ab</td>
</tr>
</tbody>
</table>
Under medium P treatment, the increment was the highest for all of the organs in high P efficiency soybean, with an average increment of 54.1% ($P<0.05$). Under high P treatment, there was an average increment of 69.7% ($P<0.01$) for the other organs in low P efficiency soybean, except for leaves.

Under different P concentration treatments, there were no significant effects on P distribution ratios among stems, leaves, and seeds in the high efficiency cultivars. However, compared to the other two P treatments, there was a significant increment under low P treatment. Leaves and pod shells were mostly influenced under medium and high P treatments in low P efficiency soybean. Under low P concentration, compared to the low P efficiency soybean, the P accumulation of leaves and pod shells was markedly higher in high P efficiency soybean; however, there was a significantly higher P accumulation in leaves and pod shells in high P efficiency soybean than those in the low ones. Under low P treatment, P was mainly allocated to leaves in high P efficiency cultivars, but the allocation was similar in the leaves and stems in low P efficiency cultivars. Under medium P and high P treatments, P was primarily allocated to stems in high P efficiency soybean. Additionally, in low P efficiency cultivars, P was dominant to leaves under medium P treatment, and to stems and pod shells under high P treatment.

**Mature stage** Under different P treatments, there was no significant effect on P content of various organs in high P efficiency soybean at the mature stage, but there was a significant improvement in stems and leaves at medium and high P treatments compared with low P treatment (Table 5). The increments of P content in stems and leaves were higher under medium P treatment (90.0 and 57.9%) than those under high P treatment (40.0 and 31.6%).

The accumulation of P in stems and leaves of high P efficiency soybean showed a significant increment under medium and high P treatments, compared with low P treatment. Besides, the accumulation of P in all organs was significantly increased in low P efficiency soybean.

Under low P treatment, the P contents in stems and leaves were significantly much higher in high P efficiency soybean than those in low P efficiency soybean. Under both low and high P treatments,
P accumulation in stems, leaves and seeds was significantly higher in high P efficiency soybean than those in low P efficiency soybean. Under moderate P conditions, distribution of P in stems and leaves was lower in high P efficiency soybean than that in low P efficiency soybean.

Effect of P on dry matter accumulation and P utilization rate in various P efficiencies soybean cultivars

**Biomass** There was little difference in biomass between the high and low P efficiency cultivars at the early pod-filling stage under low P treatment (Fig. 1), while a significant difference was existed at the late pod-filling stage, increased by 23.1% in average. Under moderate P concentrations, there was little difference among the two types of cultivars. Under high P treatment, biomass was significantly much higher in high P efficiency soybean than that in low P efficiency soybean, increased by 17.4% in average, in addition, difference between the two type cultivars increased after the pod setting stage. Under medium and high P treatments, the biomass of high P efficiency cultivars increased by 18.8 and 16.0% in average, respectively; while this increased by 41.0 and 40.1%, respectively, at the grain-filling stage. In addition, under medium and high P treatments, the average growth rates of the low P efficiency cultivars were similar, with an average of 24.5 and 25.9%, respectively. The maximal growth rate was 34.5% at the early mature stage under moderate P treatment, while was 50.8% at the blooming stage under high P treatment.

The percentage of total biomass represented slightly in each organ between the two types of cultivars (Fig. 2). Under moderate and high P levels, the percentage of biomass in stems promoted at the branching stage, which in leaves declined at the same growth stage. Under medium P treatment, the percentage of biomass in stems promoted significantly. At the blooming stage, the percentage of biomass in leaves increased under moderate and high P treatments in high P efficiency cultivars, while this value increased in low P efficiency cultivars only response to high P treatment. At the grain filling stage, the percentage of pod weight in biomass of high P efficiency cultivars increased under moderate and high P treatments, which in low P efficiency cultivars decreased under high P concentration, and the ratio of stems and leaves increased. The low P efficiency cultivars would delay mature at high P concentration, and product fewer pods. At the late growth stage, although the differences in P concentration had little influence on the percentage of seeds and pods in the two types of cultivars, different P treatments still had a great effect on stems and leaves.

**P efficiency of soybean with different P efficiencies**

P agronomic efficiency (PAE), P apparent utilization

![Fig. 1 Biomass of soybeans with various P efficiencies. A, low P treatment. B, medium P treatment. C, high P treatment. The same as below. V7, branch; R1, bloom; R3, podding; R5, grain filling; R6, late stage of grain filling; R7, early stage of maturity; R8, maturity stage. The same as below.](image-url)
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Efficiency (PAUE) and P harvest index (PHI) were much higher in high P efficiency soybean under medium P treatment than those under low and high P treatments, showing that soybean with high P efficiency had an efficient utilization of P, and fewer demand of P converted to a higher grain yield (Table 6). PHI was much higher in low P efficiency soybean under high P treatment than that under medium P treatment, indicated that low P cultivars needed more P for higher grain yield.

DISCUSSION

P was translocated and distributed easily in plant (Marschner et al. 1996). The P cycle and redistribution were very important to the improvement of growth and nutrient utilization efficiency in plants. This was especially obvious in plants under nutrient stress conditions (Marschner et al. 1996, 1997). Distribution and translocation of P was not only closely related to metabolism and the transfer of growth center, but it was also affected by P levels.

Under phosphate deficiency stress, P was transported to the regenerated tissue, which caused the plant to maintain regular growth and improved P re-utilization efficiency. Different growth stages had different requirements for P, the nutrition mechanism of P deficiency tolerance was diverse among different growth stages. Duan et al. (2002) analyzed the absorption, transportation and re-utilization efficiency of P in rape at various stages. It was found that roots had a relatively strong absorptive capacity for P at the

<table>
<thead>
<tr>
<th>Treatment</th>
<th>P agronomic efficiency (kg kg⁻¹)</th>
<th>P apparent utilization efficiency (%)</th>
<th>P harvest index (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>High P efficiency</td>
<td>Low P efficiency</td>
<td>High P efficiency</td>
</tr>
<tr>
<td>Medium P</td>
<td>4.9 a</td>
<td>1.7 a</td>
<td>9.7 a</td>
</tr>
<tr>
<td>High P</td>
<td>1.7 b</td>
<td>1.3 a</td>
<td>4.8 b</td>
</tr>
</tbody>
</table>

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seedling and bud stages, and normal development of seeds was ensured by the sustainable and strong re-use of P after the blooming stage.

In present, under P deficiency, 68.4% of P was supplied to leaves in high P efficiency cultivars, while 62.7% in low P efficiency cultivars at the branching stage. Besides, the distribution ratio of P increased in stems of the high P efficiency cultivars, while P was still supplied to the leaf of low P efficiency cultivars. Under low P condition, the P supplement to stems and leaves was equivalent in high P efficiency cultivars with 49.7 and 50.3%, respectively; while P was distributed mainly to leaf, 55.7%, and less in stems, 44.3%, in low P efficiency cultivars. The content of P was lower in seeds of high P efficiency cultivar at the end of pod-filling and mature stages, with 22.5 and 69.8%, respectively; while relative higher in low P efficiency cultivars at the same growth stages, with 26.0 and 74.2%, respectively. Our results showed that the high P efficiency cultivars had a great capacity to coordinate and re-use P. This ensured that these cultivars had a high P content under low P conditions, assuring an adequate rate of photosynthesis and dry matter production, which meant they used a relatively small amount of P to produce a higher grain yield.

Dry matter and nutrient accumulation were the basis of organ differentiation and yield formation, and nutrient absorption benefited the formation and accumulation of dry matter. Dry matter and nutrient accumulation were susceptible to P in the whole growth period of soybean. P effected the development and symbiotic nitrogen fixation of soybean, appropriate P levels improved dry matter accumulation, and then greatly increased grain yield (Hu et al. 2002; Dong 2009). Under suitable P concentrations, dry matter accumulation was distributed more strongly in the podd resulted in an improvement of yield. Therefore, dry matter production and changes in the distribution ratios of different organs were the critical factors in restriction of yield (Gourley et al. 1994; Cai et al. 2004).

Our study demonstrated that under moderate and high P concentrations, although the low P efficiency cultivars had a strong rate of the dry matter increment, which in two type cultivars increased slightly, especially in high P efficiency cultivars. Low P stress had little effects on the growth of low P efficiency cultivars, due to their excellent photosynthetic rate and carbon assimilation of these cultivars (Cao and Pan 2000; Zhang et al. 2006). In addition, the P accumulation level of whole plants in high P efficiency cultivars increased by 24.4% than that in low P efficiency cultivars in average, and this rate was similar in various soybean organs. The percentage of P was higher in organs of high P efficiency cultivars. Under moderate and high P levels, the high P efficiency cultivars were little influenced, exhibiting excellent PAE (Table 6) and PHI. PAUE was defined as the P uptake per P fertilization, and this value was much higher in high P efficiency cultivars than that in low P efficiency cultivars, which explained the reason that P content of whole plants in high P efficiency cultivars was slightly influenced by P. The whole plants and various organs of high P efficiency cultivars had relative high P levels, which ensured normal metabolism, leading to an increment in biomass.

MATERIALS AND METHODS

Plant materials

A total of 226 soybean (Glycine max) cultivars (lines) were previously evaluated for the ability of tolerance to P stress (Li et al. 2004). Of these, Jindou 33 and Liaodou 13 were chosen as high P efficiency cultivars (lines), while Tiefeng 3 and Jin 8-14 were chosen as low P efficiency cultivars (lines).

Research design

Field experiments were conducted at the research farm of Shenyang Agricultural University, and physiological index were measured at the key laboratory of crop physiology, ecology, genetics and breeding, Ministry of Agriculture. The soil test indicated that the total nitrogen content was 2.0 g kg\(^{-1}\), with a total P content of 87 mg kg\(^{-1}\), an available nitrogen level of 74.3 mg kg\(^{-1}\), an available P level of 1.7 mg kg\(^{-1}\), an available potassium level of 149.4 mg kg\(^{-1}\), and an organic matter content of 18.96 g kg\(^{-1}\), pH 5.4. The experimental design was a split plot arrangement in three different concentrations of P (P pentoxide) with three biological replicates, included 82.5, 165 and 0 kg ha\(^{-1}\) as control. The plots were 5 m long by 3 m wide (five rows), with a final density of 150 000 plants ha\(^{-1}\) (0.11 m plant spacing). The plants were planted on May 1st, harvested on October 1st, 2009 and 2010, and were grown under conventional management.
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Methods

Four plants were sampled at the branch, bloom, pod-setting, seed filling, final seed filling stages, and 10 plants were harvested at the mature stage, to determine the dry matter weight of the above ground parts and all organs. The harvested material was divided into stem, leaf, pod, shell, and seed, followed by drying, grinding, and then made analyses. Each sample was digested using the H2SO4-H2O2 method, followed by the measurement of total nitrogen content using the Kjeldahl method. The vanadium molybdate yellow colorimetric method was employed to determine the P content, and atomic absorption spectroscopy was used to measure the potassium concentration (Bao 2005). The percentage content of P per dry matter weight of each organ represented the P accumulation rate.

P fertilizer use efficiency was represented by PAE, PAUE and PHI (Huggins and Pan 1993; Lopez-Bellido and Lopez-Bellido 2001; Wen et al. 2003). PAE (kg kg⁻¹)=(W1-W2)/G; PAUE (%)=[(P1-P2)/G]×100; PHI (%)=[(P1_w-P2_w)/(P1-P2)]×100

Where, W1, yield of seed under P treatment; W2, yield of seed under no P treatment; G, P application amount; P1, the amount of absorbed P in aboveground part under P treatment; P2, the amount of absorbed P in aboveground part under no P treatment; P1_w, the amount of absorbed P in seed under P treatment; P2_w, the amount of absorbed P in seed under no P treatment.

Data analyses

SPSS15.0 and Excel 2003 were used for statistics and analyses. All data were averaged over a 2-yr-period.

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References


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