

# Genetic Dissection of Low Phosphorus Tolerance Related Traits Using Selected Introgression Lines in Rice



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**Abstract:** To dissect the genetic basis of low phosphorus tolerance (LPT), 114 BC<sub>2</sub>F<sub>4</sub> introgression lines (ILs) were developed from Shuhui 527 and Minghui 86 (recurrent parents), and Yetuozei (donor parent). The progenies were tested for 11 quantitative traits under three treatments including normal fertilization in normal soil (as control), normal fertilization in barren soil and low phosphorus stress in barren soil in Langfang, Hebei Province, China. Moreover, the ILs were investigated at the seedling stage using nutrient solution culture method in greenhouse in Beijing, China. A total of 49 main-effect quantitative trait loci (QTLs) underlying yield related traits were identified in Langfang, and their contributions to phenotypic variations ranged from 6.7% to 16.5%. Among them, 25 (51.0%) QTLs had favorable alleles from donor parent. A total of 48 main-effect QTLs were identified for LPT-related traits in Beijing, and their contributions to phenotypic variations ranged from 7.7% to 16.6%. Among them, 21 (43.8%) QTLs had favorable alleles from donor parent. About 79.6% of the QTLs can be detected repeatedly under two or more treatments, especially QTLs associated with spikelet number per panicle, spikelet fertility and 1000-grain weight, displaying consistent phenotypic effects. Among all the detected QTLs, eight QTLs were simultaneously identified under low phosphorus stress across two environments. These results can provide useful information for the genetic dissection of LPT in rice.

**Key words:** rice; phosphorus tolerance; yield; introgression line; quantitative trait locus

Rice (*Oryza sativa* L.) is the staple food for more than half of the world's population (Huang et al, 2010). The developments of semi-dwarf rice varieties in the 1960s and hybrid rice varieties in the 1970s brought great increase in average yield, which have dramatically contributed to the self-sufficiency in world rice supply (Ma and Yuan, 2015). Recently, the rapid population growth and economic development have imposed a heavy pressure on rice production. To meet the global rice requirement, the total rice production needs to be increased by 0.6%–0.9% per year (Carriger and Vallee, 2007; Vinod and Heuer, 2012). It is well known that the fertilizer input plays an important role in improving yield. However, the contribution of fertilizer input to

rice yield increase has gradually decreased in the past ten years (Wu, 2013). The overuse of fertilizer not only causes the rising agricultural energy consumption, but also results in environmental degradation and pollution (Wu, 2013; Huang et al, 2014). Phosphorus (P) is a critical nutrient for rice growth and development, and it is also a basic component of many organic molecules, especially nucleic acids and proteins (Lea and Mifflin, 2011). P deficiency leads to the lack of nutrition and the lag of plant growth (Guo et al, 2013). Besides, P deficiency is usually the secondary factor for low pH soils, which imposes restrictions on root growth although there are high concentrations of iron and aluminium (Ismail et al,

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2007). Moreover, it causes a series of molecular and physiological responses that ultimately result in significant yield loss in rice (Dobermann and Fairhurst, 2000; Ismail et al, 2007). Currently, P deficiency occurs to about 50% of the agricultural soils in many Asian, African and South America countries (Lynch, 2011). Therefore, the balanced and sustainable use of P fertilizer is of paramount importance (Vinod and Heuer, 2012). One of the most effective ways to ensure the sustainable development of agriculture is to develop and grow rice varieties with low P tolerance (LPT) or high P-use efficiency.

In recent years, molecular-marker technology has contributed to the development of genetic map that makes it possible to identify quantitative trait loci (QTLs)/genes related to LPT in rice, including P absorption and utilization. However, very few QTLs can be directly applied in rice breeding (Gao et al, 2006; Mu et al, 2008; Vinod and Heuer, 2012; Wu, 2013), which is largely due to the complex genetic basis of LPT and the lack of QTL validation in different genetic backgrounds and environments (Loudet et al, 2003). Based on the advanced backcross QTL analysis (Tanksley and Nelson, 1996), Li (2005) proposed a molecular breeding strategy that combines QTLs/genes mining with target trait improvement using selected introgression lines (ILs) (Luo, 2005). The selected ILs were derived from elite varieties (recurrent parents) and can be directly applied in the practical breeding and QTL discovery. Research regarding QTL detection in selected ILs has been reported, especially for drought, salt and diseases (Zheng et al, 2007; Zang et al, 2008; Chen et al, 2011). However, systematic identification of the QTLs for LPT has not been conducted in a large scale.

Therefore, 114 BC<sub>2</sub>F<sub>4</sub> ILs in two populations deriving from three parents, Minghui 86 (recurrent parent), Shuhui 527 (recurrent parent) and Yetuozai (donor parent), were selected to investigate the genetic basis for LPT both at the seedling and maturity stages of rice. The phenotypic evaluation and QTL identification of LPT-related traits in ILs were also conducted. This study will probably provide better understanding of LPT and useful information for marker-assisted selection in rice breeding.

## MATERIALS AND METHODS

### Population development

Two widely used elite indica restorer lines (Shuhui

527 and Minghui 86) were used as recurrent parents to develop two backcross populations with Yetuozai (donor parent), an indica rice variety. The F<sub>1</sub> combinations were backcrossed with the corresponding recurrent parents to generate BC<sub>1</sub>F<sub>1</sub> seeds, and 25 random BC<sub>1</sub>F<sub>1</sub> plants were backcrossed with the recurrent parents to obtain 25 BC<sub>2</sub>F<sub>1</sub> lines. All BC<sub>2</sub>F<sub>1</sub> lines were planted in the fields and allowed to self-produce BC<sub>2</sub>F<sub>2</sub> seeds which were bulk-harvested as a random BC<sub>2</sub>F<sub>2</sub> population. A total of 1000 plants from each BC<sub>2</sub>F<sub>2</sub> population were screened randomly in a low P red soil field without P fertilizer input at the experimental station of Xuancheng Institute of Agricultural Science, Anhui Province, China, in the summer season (May to September) of 2010, and 60 and 54 ILs were selected from Shuhui 527/Yetuozai (SY) and Minghui 86/Yetuozai (MY) populations, respectively. Then, 114 BC<sub>2</sub>F<sub>4</sub> lines were developed from self-crossed generations at the experimental station of the Institute of Crop Science, Chinese Academy of Agricultural Sciences in Sanya the winter season (November of 2010 to April of 2011).

### Phenotypic evaluation

The phenotypic evaluation was conducted under field conditions in Langfang, Hebei Province and nutrient solution culture conditions in Beijing, China.

#### *Field conditions*

The phenotypic evaluation under field conditions was conducted in the summer of 2013 at the experimental field of the International Agricultural High-Tech Industrial Center of the Chinese Academy of Agricultural Sciences (Langfang, China). Three treatments included normal fertilization in normal soil (treatment A, as control group), normal fertilization in barren soil (treatment B) and low P stress in barren soil (treatment C). The 50 cm topsoil was replaced with uniform barren soil except the plot of the control group. The organic matter content, total contents of nitrogen, phosphorus and potassium in the soil are listed in Table 1. All 114 BC<sub>2</sub>F<sub>4</sub> lines and 3 parental lines with three treatments were planted under a randomized block design. Each treatment consisted of three replications (plots), and each IL was in one row with 12 plants. The seeds of each IL were sown in the seedbeds in late April. Then the seedlings were transplanted to experimental fields on 6 June with a spacing of 25.0 cm between rows and 17.0 cm within each row. Fertilization was conducted in two phases, the stem elongation stage (11 July) and booting stage

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