

## Reducing Phytic Acid in Rice Grain by Targeted Mutagenesis of a Phospholipase D Gene

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**Abstract :** Phospholipids are one of the major classes of lipid comprising 10% of total grain lipid in rice. Phospholipids are the main phosphorus containing lipid in the rice endosperm, contributing to rice palatability and seed storage property. However, in the rice grain, the majority of phosphorus occur in the form of phytic acid and are highly abundant in the bran. Phytic acid, also known as hexaphosphorylated inositol (IP6), are strong chelating agents which reduces the bioavailability of essential dietary nutrients and are therefore less desirable by rice breeders. We used the CRISPR/Cas9 system to generate mutants of a phospholipase D gene (PLD $\alpha$ 1), which is responsible for the degradation of phospholipids into phosphatidic acid (PA). In the mutants, we found a significant reduction in the concentration of phytic acid in the grain as compared to the wild-type. The biochemical analysis of the PLD $\alpha$ 1 mutants showed that the decrease in production of phosphatidic acid is due to reduced accumulation of CDP-diacylglycerol-derived phosphatidylinositol (PI), ultimately leading to lower accumulation of phytic acid in mutants. These results showed that loss of function of PLD in rice leads to lower production of phytic acid, suggesting the potential application of Ospld $\alpha$ 1 in breeding rice with less phytic acid.

**Keywords :** CRISPR/Cas9, phospholipase D, phytic acid, rice

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