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Enhancing phosphorus use efficiency of rice with
P-localized application in P-deficient paddy field

Phosphorus is crucial for plant growth, especially in rice, a vital crop in Sub-Saharan Africa. However, phosphorus deficiency in soil is a significant challenge, limiting crop yield. Conventional application of fertilizer through broadcasting is often ineffective due to phosphorus fixation in soil and the limited purchasing capacity of farmers. Recent studies suggest phosphorus localized application, such as P-dipping, as a cost-effective alternative. P-dipping involves dipping rice seedling roots in a phosphorus-enriched slurry before transplanting them into the paddy field. This method has shown promising results in increasing biomass growth and yield while reducing phosphorus application rates. However, it also presents challenges such as the risk of salt stress and transplanting shock of seedlings, particularly when phosphorus concentration is high and in high-temperature environments. Despite these challenges, P-dipping offers potential benefits, including improved resilience to submergence stress and shortened growth durations. Shortening the rice growth duration is particularly beneficial in fields where rice is often exposed to late-season cold stress. However, limited knowledge exists regarding the role of root volume and root morphology in the efficacy of this technique. We hypothesized that the variability of the root volume of seedlings may impact the amount of P-slurry adhered to roots. Therefore, our study aims to determine the optimal seedling age for maximizing biomass and yield effects from P-dipping. Additionally, further research is needed to understand the synergy effect of the P-dipping and root phenotypes of specific genotypes for optimal application.

In our first study, we assume that a higher root volume of older seedlings may lead to an increase in P-slurry. However, old seedlings with low vigor pose risks of transplanting shock, exacerbated by fertilizer burning from high P concentration of P-dipping. On the other hand, young seedlings recover faster potentially mitigating these risks, but have relatively small root volume for significant transfer of the phosphorus slurry to the main field. Therefore, our pot experiments implying various seedling ages were conducted to investigate the change of P-amount adhered to various seedling ages. Our results identified that the amount of slurry from P-dipping increased not linearly but in a sigmoidal pattern against seedling age, plateauing around 6.0 seedling age. Correspondingly, the effect of P-dipping on initial biomass growth was maximized around the 6.0 seedling age and then was reduced when combined with much older seedlings, attributable to the prolonged period of transplanting shock. Similarly, on-farm trials among 90 farmers' fields in the central highlands of Madagascar were conducted to compare control (no P fertilizer) and P-dipping treatment. After categorizing the seedling age into three groups of young (< 4.5 seedling age), intermediate (4.5–6.3 seedling age), and old (> 6.3 seedling age), our results reported the highest biomass from P-dipping at 40 days after transplanting when combined with the intermediate seedling age group. Furthermore, the yield gain of P-dipping (yield difference from P-dipping to control) was also higher with intermediate seedling age than the young and old seedlings group. It suggests the beneficial effect of using vigorous seedlings around 6 leaves with P-dipping. Specifically, 6-leaf seedlings have a higher root volume for adhering to P-enriched slurry than younger seedlings, while also having a lower risk of transplanting shock than older seedlings.

Furthermore, we conducted a comprehensive pot experiment to investigate the effects of P-dipping compared to P incorporation (which used 4 to 15 times more P) on the morphological growth and component yield across different seedling ages. We tested three groups of seedlings, aged 2.9 ± 0.1 leaves, 4.1 ± 0.2 leaves, and 6.5 ± 0.2 leaves. Each group was subjected to three different treatments: P-dipping, P incorporation, and control without P treatment. The results highlighted the dynamic relationship between phosphorus application methods and seedling age. For the youngest seedlings of 2.9 leaves, P incorporation had a more substantial effect on overall plant growth (morphological growth and component yield). However, as the seedlings aged, the pronounced effects of P incorporation diminished. The difference was more or less different at 4.1 leaves. By the time the seedlings reached 6.5 leaves, no significant differences in growth parameters were observed between the P-dipping and P-incorporation treatments. This confirms our previous finding that approximately 6 leaf age had a greater response with P-dipping even compared to P incorporation. Notably, at harvest, the aboveground biomass and panicle weight showed greater benefits from P-dipping in the older seedlings with 6.5 leaves. These findings provide valuable insights for smallholder farmers on optimizing P fertilizer application based on the age of their seedlings in the nursery. This nuanced approach to phosphorus management can lead to more efficient use of phosphorus fertilizers, cost savings, and improved crop performance.

In our other study, we focused more on the root morphology change induced by the P-localized application via P-dipping. In our first experiment, we investigated two P placements at the surface (5 cm) and at the deep (20 cm), each under a continuously waterlogged system and moderate soil moisture content of 30%. We hypothesized that P-dipping would be more advantageous at the surface due to vigorous root development in the topsoil, but less effective with deeper placement, especially under moderate soil moisture conditions. However, our results suggested that the effect of both P placements did not interact with soil moisture conditions. Therefore, P-dipping placement at 5 cm depth tends to have higher growth parameters than P-dipping placement at 20 cm depth, whether under a continuously waterlogged system or moderate soil moisture conditions. Specifically, higher root development of nodal root and L-type lateral root was observed in the topsoil layer with P placement at the surface, especially under a waterlogged system. On the other hand, the root distribution was more and less uniform with deeper P placement on both soil moisture conditions. These results suggest the crucial role of adequate water and the strategic localization of P hotspots from P-dipping at the surface for maximizing its efficiency.

In the second part of our study, we conducted a pin-board experiment involving a mutant genotype called *our1*, and its wild type. The mutant carries the *our1/osbzip1* gene, responsible for promoting the growth of thicker L-type lateral roots while repressing thin S-type lateral roots initiation. We compared P-dipping treatments against uniform P application (P incorporation). Our findings revealed that P-dipping induced higher P contents at 35 days after transplanting in the wild type compared to the *our1* mutant. Furthermore, the P-dipping increased significantly the S-type lateral root length and the nodal root length in the wild type, particularly in the upper soil layer (0−12 cm) where the P hotspot from P-dipping was localized. By investigating the relationship between P uptake change by P-dipping and root length change by P-dipping between the two genotypes, we noted that S-type change was more significant than nodal root change for higher P uptake. This suggests that the proliferation of S-type lateral roots plays a crucial role in enhancing P uptake from P-dipping. This result provides additional understanding for optimizing the P-dipping technique for higher rice productivity in Sub-Saharan Africa.

Our study highlights the efficacy of P-dipping for addressing phosphorus deficiency in rice cultivation. Relatively older seedling age, around 6.0 leaf age, optimizes biomass and yield

effects while mitigating transplanting shock, leading to increased initial biomass and subsequent grain yield. Furthermore, the proliferation of S-type lateral roots and nodal roots induced by P-dipping significantly enhances phosphorus uptake. These findings emphasize the importance of seedling age and root morphology at transplanting in maximizing the benefits of P-dipping for sustainable rice production in phosphorus-deficient soils. Further research into the genetic mechanisms underlying root responses and synergetic effect on other nutrients with localized phosphorus application could offer valuable insights for enhancing crop resilience and sustainable rice productivity.