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Fe uptake and translocation responded to various Zn supplies at tillering stage: Coupling study with stable metal isotope and molecular biology

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Metal stable isotope is now emerging as a powerful technique to study metal uptake and translocation mechanisms in plants. Recent studies observed Fe homeostasis regulated by iron-regulated metal transporters contributing to the tolerance of excess Zn, but limited effort has been invested in understanding competitive interactions between Zn and Fe with biological properties and metal stable isotope in rice. Here, we investigated isotopic compositions of iron (Fe) during uptake from nutrient solutions by rice (*Oryza sativa* L) at different Zn supplies to explore Fe uptake and translocation mechanisms. The results show that tillering stage rice was more enriched in heavy Fe isotopes with the increase of Zn concentration. The average isotope fractionations of the rice plant as a whole, relative to solution ($\Delta^{56}Fe_{plant-solution}$), were -0.45‰ and 0.01‰ at sufficient Zn (1 μM) and excess Zn (100 μM) treatment, respectively.

Rice directly absorbs Fe^{2+} with the Strategy I system regulated by Fe^{2+} transporter gene *OsIRT1*, while absorbs Fe^{3+}-Ps with the Strategy II system regulated by phytosiderophore efflux transporter gene *OsTOM1* and Fe-Ps transporter gene *OsYSL15*. The different fractionation factors of iron stable isotopes were found to be resulted from the expressions of different transporter genes for Fe translocation in rice when with different Zn supplies. Real-time quantitative PCR analysis revealed that with the increase of Zn concentration, the expression of *OsIRT1* was down-regulated, while the expression of *OsTOM1* and *OsYSL15* were significantly up-regulated. The expression patterns indicate that rice absorbs more Fe^{3+}-Ps with Strategy II with the increase of Zn concentration, in which fewer iron redox reactions occurred during the translocation and resulted in decreased Fe isotope fractionation factors ($\Delta^{56}Fe_{root-shoot}$ from 2.42‰ to 1.62‰). The results demonstrate that Fe isotope fractionation and gene expression analysis could provide insight into the internal multimetal transport pathways, further clarify the interaction mechanism of Fe and Zn uptake and translocation during rice growth.