

Analysis of Mg ion transport by AtMRS2-1 in *Arabidopsis thaliana* using Mg-28

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The maintenance of intracellular Mg concentration is primarily controlled by Mg transporters. The Mg²⁺ transporters in *Arabidopsis thaliana* have been extensively studied. Current studies suggest that Mg transport across plant membranes involves the influx-type MRS2/MGT family,¹⁾ the efflux-type MGR family,²⁾ and other divalent cation transporters. However, the specific roles of individual transporters are not yet fully understood.

Within the MRS2/MGT family, the present study focused on AtMRS2-1, a vacuole-localized Mg transporter. AtMRS2-1 knockout mutants and complemented lines expressing AtMRS2-1 under its native promoter were developed to allow detailed investigation. The analysis demonstrated growth alterations in the AtMRS2-1 knockout lines under varying Mg conditions.

This study aimed to elucidate the role of AtMRS2-1 in Mg uptake and transport within plants. Using AtMRS2-1 mutant and complemented line, Mg-28 tracer experiments were performed to investigate its involvement in Mg dynamics, providing new insights into its physiological function.

The Mg transport properties of wild-type plants (WT), AtMRS2-1 knockout mutants (*mrs2-1*), and newly generated complementation lines (*mrs2-1* complemented with the AtMRS2-1 promoter and genomic cDNA; referred to as Com) were analyzed using Mg-28 produced in the ²⁷Al(α , 3p)²⁸Mg reaction at the RIKEN AVF cyclotron in May 2024. Plants grown on agarose gel were used for the experiments. Mg-28 was supplied to a 1 cm section of the root for 2 hours, followed by an additional 2-hour period for redistribution. The distribution of Mg-28 within the plants was then quantitatively analyzed using an imaging plate (Fig. 1).

The results showed that, compared to WT and Com plants, *mrs2-1* mutants exhibited a higher proportion of Mg-28 retention rate in the absorbed root section. This suggests that AtMRS2-1 plays a role in Mg mobilization within the roots.

We hypothesized that Mg absorbed by the roots is partially stored in vacuoles during its passage through root cells, remaining within the root tissue. AtMRS2-1 is considered to function as a transporter that remobilizes Mg from vacuolar stores back into the cytoplasm. This remobilization of vacuolar Mg into the cytoplasm is critical for maintaining plant growth.

To test this, we cultivated WT, *mrs2-1*, and Com plants under Mg-excess conditions. The results showed that *mrs2-1* mutants showed better growth compared to WT and Com plants. This suggests that, under Mg

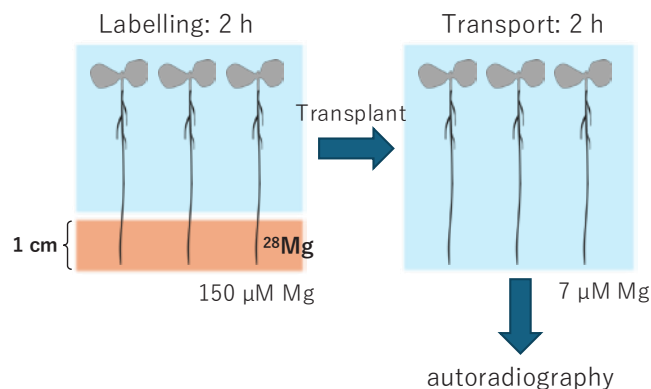


Fig. 1. Experimental Method. After a 2-hours absorption of Mg-28, samples were transferred to Mg-free medium (containing 7 μ M Mg) and cultured for an additional 2 hours. The Mg-28 signal was detected using autoradiography.

Mg retention rate at root tip (1 cm)

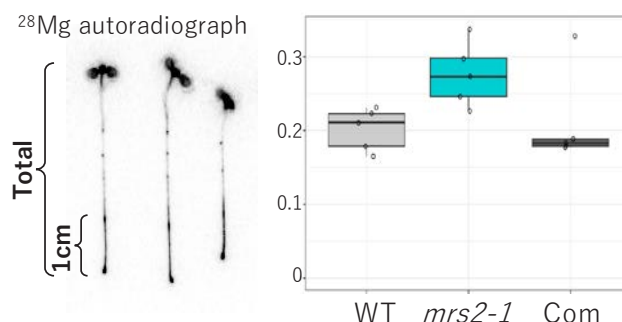


Fig. 2. The retention rate of Mg-28 in the apical 1 cm of the root after a 2-hours absorption period followed by an additional 2-hours cultivation in Mg-free medium (containing 7 μ M Mg). $n = 4$.

excess conditions, it is better for the plant not to have AtMRS2-1, which moves Mg ion into the cytoplasm.

These findings suggest that fine-tuning how AtMRS2-1 works could help *Arabidopsis thaliana* tolerate changes in environmental Mg levels. To explore this, we are currently studying molecules that might control AtMRS2-1's Mg transport activity.

Acknowledgement

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References

- 1) M. Gebert *et al.*, Plant Cell **21**, 4018 (2009).
- 2) S.-F. Meng *et al.*, Mol. Plant **15**, 805 (2022).

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