

OA-05

## Regulatory Roles of Glutamate in Drought Stress Tolerance in *Brassica napus*

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### [Introduction]

As global warming becomes more serious, drought stress is becoming more frequent and severe which negatively affects crop productivity and quality. Drought induces imbalance between reactive oxygen species and antioxidant systems, resulting in oxidative stress which induces changes in redox status and phytohormones (La et al., 2019; Park et al., 2022; Lee et al., 2022a). Several studies have reported that proline and glutathione are related to drought stress tolerance by regulating the synthesis and signaling of phytohormones (La et al., 2020; Lee et al., 2022b). However, the role of glutamate, a precursor of proline and glutathione, in the drought tolerance mechanism is not well understood.

### [Materials and Methods]

*Brassica napus* (cv. Mosa) was grown in the greenhouse condition. To clarify how glutamate pretreatment respond to drought stress, experiments were conducted for long-term and short-term periods. Plants were pre-treated with 5.5 mM glutamate treatment for 18 or 2 days during the long-term or short-term periods, respectively. Glutamate-pretreated plants were irrigated with 20 ml of water to induce drought stress for 14 or 4 days during the long-term or short-term periods, respectively. Non-treated group (control) was normally irrigated with 200 ml for well-watered plants and 20 ml for drought-stressed plants. The collected leaf-sample were immediately frozen in liquid nitrogen and stored in a freezer (-80°C) for further analysis of metabolites, redox balance, gene expression and hormone levels.

### [Results and Discussion]

Drought stress-induced H<sub>2</sub>O<sub>2</sub> accumulation was significantly alleviated by glutamate pretreatment for long-term or short-term periods. The accumulation of ammonia in drought-stressed plants coincided with the upregulation of *glutamate dehydrogenase 2* and *glutamate dehydrogenase 3* gene expression, leading to the reduction of nitrogen assimilation-related genes, *glutamine synthetase 2 (GS2)* and *ferredoxin-glutamate synthase 1 (Fd-GOGAT1)* for a long-term period. Glutamate pretreatment under drought stress largely enhanced expression of *GS2* and *Fd-GOGAT1*, and reduced ammonia accumulation, accompanied by a less induction of proline content. These results indicate that glutamate alleviates drought stress by activating nitrogen assimilation and reducing ammonia accumulation. In addition, glutamate pretreatment alleviated decreasing of pool size of reduced glutathione, which plays a role in removing ROS and redox balance, resulting decrease drought stress symptoms for a short-term period. The upregulation of abscisic acid (ABA) synthesis-related gene 9-cis-epoxycarotenoid dioxygenase 3 and ABA signaling-related gene ABA insensitive 5, known as a stress hormone, was mitigated by glutamate pretreatment. Taken together, these results suggest that glutamate enhances drought stress tolerance by modulating nitrogen assimilation and glutathione synthesis in relation to ABA metabolism.

### [Acknowledgement]

This work was supported by a grant from the National Research Foundation of South Korea under project NRF-2022R1I1A3072357.

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