Enhancement of ozone resistance by adjusting the stomatal aperture on leaf surface

Tropospheric ozone (O_3) is a major photochemical oxidant and one of the most phytotoxic air pollutants. Ozone levels have been increasing in many parts of the world, with high concentrations of ozone causing serious damage to crop production. Because ozone enters the plant through the stomata, modulation of stomatal movement using transcription factors, which act as master regulators of various cellular processes, may be a useful strategy for conferring ozone resistance. However, transcription factors modulating stomatal movement have not been well characterized.

In this report, we screened a set of transgenic *Arabidopsis* lines expressing chimeric repressors for *Arabidopsis* transcription factors to identify new transcription factors related with ozone stress resistance. We found that lines expressing the chimeric repressors for GOLDEN 2-LIKE1 (GLK1) and GLK2, which have known functions in chloroplast development, exhibit remarkable ozone resistance and a closed-stomata phenotype. In addition to ozone resistance, these plants also exhibited resistance to sulfur dioxide, an oxidative stress reagent similar to ozone. On the other hand, plants that overexpress GLK1/2 exhibited higher sensitivity to ozone and sulfur dioxide, and an open-stomata phenotype. These results suggest that GLK1/2 affect ozone and sulfur dioxide resistance through the regulation of stomatal movement. We showed that lines expressing the chimeric repressors for GLK1 had reduced expression of the genes for inwardly rectifying K^+ (K^+_{in}) channels and reduced K^+_{in} channel activity, which is one of the positive regulators for stomatal opening. These results indicate that GLK1/2 act as positive regulators of genes for K^+_{in} channels and stomatal opening.

Stomata play roles in the transpiration and absorption of gases in plants. Thus, the modification of stomatal movements could prove useful for improving both the efficiency of photosynthesis and the plants' resistance to air pollutants. Regulating the expression of chimeric repressors for GLK1/2 specifically in guard cells may be a useful tool for conferring resistance to air pollutants.

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Fig. 1. Sensitivity to ozone of GLK1/2-downregulated *Arabidopsis*.
A. Two-week-old plants of wild type and GLK1/2-downregulated *Arabidopsis* (*GLK1sx* and *GLK2sx*) 1d after exposure to 0.3 ppm ozone for 7h.
B. Ion leakage of wild type and *GLK1/2sx* plants. The average of three biological replicates (three plants per replicate) is shown. Error bars represent SD.

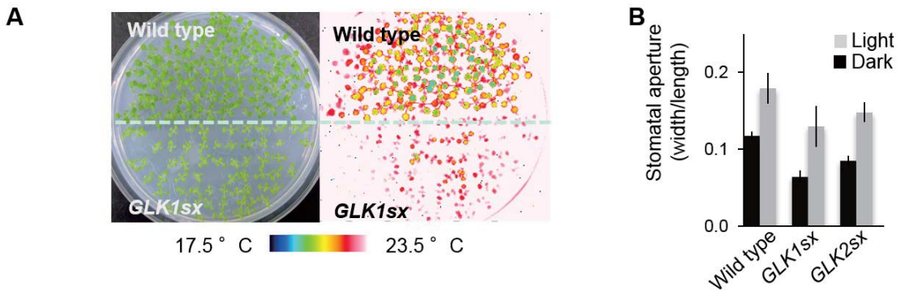

Fig. 2. Transpiration and stomatal aperture of GLK1/2-downregulated *Arabidopsis*.
A. Thermal images of wild type and *GLK1sx* plants grown on MS medium, showing the higher temperature of *GLK1sx* plants.
B. Stomatal aperture of wild type and *GLK1sx* plants grown on MS medium. The average of three independent experiments is shown (50 stomata per experiment). Error bars represent SD.