Greater Aperture Counteracts Effects of Reduced Stomatal Density on WUE: A Case Study on Sugarcane and Meta-Analysis

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Background/Objective

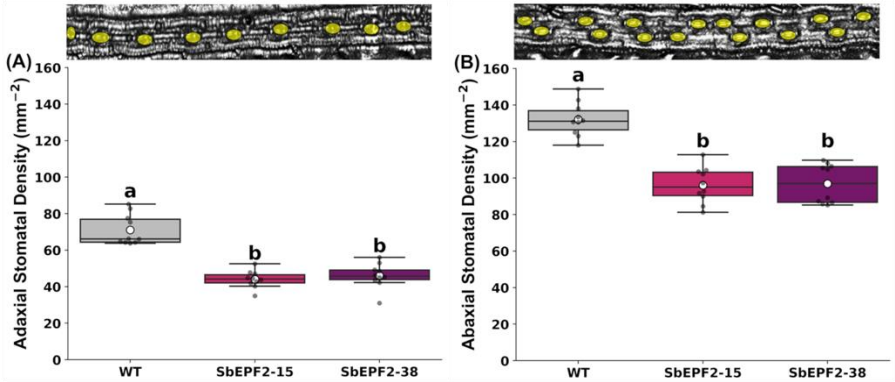
- Engineering crops to have greater water use efficiency (WUE) is a key target for crop improvement to deal with the increased frequency and intensity of drought periods resulting from climate change. At the leaf level, stomata regulate CO_2 and water vapor exchange between leaves and the atmosphere. Stomata are a target for engineering to improve crop intrinsic water use efficiency (iWUE). One example is by expressing genes that lower stomatal density (SD) and reduce stomatal conductance (g_{sw}). However, the quantitative relationship between reduced SD, g_{sw}, and the mechanisms underlying it is poorly understood.
- We addressed this knowledge gap using low-SD sugarcane (*Saccharum spp.* hybrid) as a case study alongside a meta-analysis of data from 10 species.

Approach

We aimed to (1) modulate an Epidermal Patterning Factor (EPF) gene to engineer sugarcane with lower SD and g_{sw}; (2) quantify changes in epidermal cell patterning and stomatal complex size; (3) estimate consequences of altered stomatal patterning for maximum potential g_{sw}; (4) assess changes in g_{sw} and photosynthetic physiology arising from altered SD; and (5) perform a meta-analysis to summarize the quantitative relationship between lower SD and reduced g_{sw} across C_3 and C_4 species.

Results

Transgenic expression of EPF2 from *Sorghum bicolor* (SbEFP2) in sugarcane reduced SD by 26-38% but did not affect g_{sw} compared to wildtype. Further, no changes occurred in stomatal complex size or proxies for photosynthetic capacity. Measurements of gas exchange at low CO_2 concentrations that promote complete stomatal opening to normalize aperture size between genotypes were combined with modeling of maximum g_{sw} from anatomical data.



Stomatal density of wild-type (WT) transgenic sugarcane overexpressing SbEPF2 (SbEPF2-15 and SbEPF2-38).

Significance/Impacts

This report assessed the relationship between SD and g_{sw} in sugarcane engineered to have reduced SD. Although improvements in iWUE were not achieved, the study highlighted compensatory mechanisms that operate widely across species and functional groups to diminish the degree to which reductions in SD translate into reductions in g_{sw}. These findings are important since they highlight a fundamental knowledge gap in regulating leaf gas exchange, which can be addressed to advance our efforts to improve crop iWUE.

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