

SPEED OF STOMATAL RESPONSES TO FLUCTUATING ENVIRONMENTAL CONDITIONS EXPLAINS THE LACK OF ASSOCIATION BETWEEN HEAT STRESS SENSITIVITY AND IWUE IN FIELD GROWN GRAPEVINE

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Stomata control CO₂ uptake for photosynthesis and water loss through transpiration. Phenotypic variation exists in grapevine for stomatal anatomical traits, carbon isotope discrimination (i.e. intrinsic water-use efficiency - iWUE) and heat stress (HS) sensitivity. In this work, we investigated the relationship between these traits and hypothesized that stomatal behavior to fast (i.e. minutes) environmental changes co-determines along with steady-state traits the physiological response of grapevine to the natural surrounding fluctuating environment over the growing season.

Forty genotypes of *Vitis vinifera* subsp. *sativa* and nine genotypes of subsp. *sylvestris* grown in the field were characterized for isotopic signature as well as stomatal density, stomatal size and chlorophyll fluorescence under different air temperatures. Six genotypes were then selected for contrasting traits and grown in the greenhouse. Temporal stomatal dynamics were subsequently assessed with an infra-red gas analyzer.

No relationship between iWUE, HS tolerance and stomatal traits was observed in field grown grapevine, suggesting that other physiological mechanisms are involved in determining leaf evaporative cooling capacity and the seasonal ratio of CO₂ uptake (A) to stomatal conductance (gs). Indeed, cultivars that in the field had an unexpected combination of high iWUE but low sensitivity to thermal stress, displayed a quick stomatal closure to light, but a sluggish closure to increased vapor pressure deficit (VPD) levels. This strategy aiming both at conserving water under a high-to-low light transition and in prioritizing evaporative cooling under a low-to-high VPD transition, was mainly observed in Regina and Syrah. Moreover, cultivars with different known responses to soil moisture deficit or high air VPD (isohydric vs anisohydric) had opposite behavior under fluctuating environments, with the isohydric cultivar showing slow stomatal closure to reduced light intensity but quick temporal responses to VPD manipulation. No relationship was observed between stomatal anatomical traits and time to open/close stomata, suggesting that hydraulic (fluctuations in light) and hormonal (fluctuations in VPD) signals might be involved in short-term adjustment of gas-exchange.

Our field screening provides a large physiological characterization for several traits in *Vitis* and shows the presence of a wide phenotypic variation both in *sativa* and in *sylvestris* subspecies. However, in natural field conditions, leaf overlapping and cloud cover impose fast changes in light and VPD levels; suboptimal stomatal adjustment can lead to nonsynchronous behavior between A and gs, which can result in reduced iWUE and lowered leaf evaporative cooling under high temperature conditions. We propose that stomatal behavior to fast environmental fluctuations can play a critical role on leaf thermoregulation and water conservation under natural field conditions.