



How do we work? Determination of heat tolerance

Background: During photosynthesis, the activation of chlorophyll makes electrons flow over the membrane of the chloroplast, a risky step – under heat stress, these electrons can attach to oxygen, forming Reactive Oxygen Species (ROS). These ROS attack membranes, proteins, and DNA. Under heat stress, grapevine tries to lower leaf temperature by stimulating transpiration. However, this will consume more water. Since heat usually comes with drought, the final resort out is to shed the leaves as to avoid damage to the whole plant. Some wild grapevines, but also some domesticated varieties can generate **heat shock proteins**. These form a protective layer around membranes and proteins, enabling photosynthesis also at higher temperatures, such that the plant can safeguard the water otherwise needed for cooling.

Principle of the measurement: We monitor, how the genes for these protective proteins respond to heat stress. Herefore, we cut out small leaf discs and place them on an agar plate, where they are subjected to a controlled heat shock. This not only allows to measure a large number of samples, but also improves the reproducibility of the test. After defined time points, the leaf discs are shock-frozen in liquid nitrogen to extract the RNA and measure gene expression through real-time qPCR. This allows to see, how strong different heat-shock genes are activated. Comparing the results with data from whole plants subjected to heat stress, showed that our leaf-disc assay can predict the heat tolerance of the respective grapevine variety very well, although much less time, space and labour is needed to get the result.



How do we work? Determination of heat tolerance

How it works: Using a cork borer, we excise the leaf discs and place them on agar in Petri dishes. To prevent that the wounding stimulus perturbs the response to heat, we first let the „pain“ fade out. After one day, the response of the wounding genes is completely gone. Only then, we apply the heat shock. We use 42°C, as it is happening during one of the progressively frequent hot summer days. Of course, we also run a so-called negative control that is treated in exactly the same way, just omitting the heat shock. After different time points, we measure then the expression of the Heat Shock Proteins (HSPs). Here, we investigated several types and found out that especially HSP17 and HSP18 are good predictors for heat tolerance. A strong and fast activation of these genes tells that the respective grapevine variety can cope well with heat stress. For instance, the heat susceptible Riesling induces those genes much more weakly than our wild grapevine “champion” *sylvestris* Hördt 29.

