

One of the main challenges following climate change is long, unpredictable periods of drought. In our search towards increased plant resilience against drought stress, we are studying several endophytic microbial strains conferring such drought tolerance in maize.

In this project we aim to identify their mode of action (MoA) and elucidate the downstream signalling in the maize leaf. For all species, a kinematic analysis to determine the contribution of cell division and cell elongation has already been performed. This allows us to sample our material in a strategic manner, as the cell division zone and cell elongation zone are spatially separated along the maize leaf. In a next step, we have performed a transcriptomic analysis on the maize leaf meristem of endophyte-inoculated maize plants that were subjected to drought stress. This allowed us to identify differentially expressed genes that could be relevant in the MoA of these endophytes.

One of our key findings is the upregulation of genes related to biotin biosynthesis in drought-stressed plants after inoculation with fungal strain ESALQ1307. In an experiment where we supplied drought-stressed plants with biotin, we could observe an improvement in leaf elongation rate (LER), indicating that elevated biotin levels do have a positive effect on maize growth and thus drought tolerance. We are currently screening different mutant maize lines, impaired in biotin biosynthesis, for growth effects under drought stress. In a next step we plan to quantify biotin levels in fungus-inoculated and mutant plants using UPLC-MS/MS.

Through collaborations with a Chinese university, we managed to obtain bacterial strains 327, 571, and BR5. Here too, a transcriptomic analysis allowed us to put forward a potential MoA explaining the observed improved drought tolerance. For 327, we could observe a downregulation of genes responsible for lignin biosynthesis. In a next step, we aim to study the growth of mutant maize lines, impaired in lignin biosynthesis, under drought conditions. Regarding 571, we have found an upregulation of trehalose-6-phosphate synthases (TrPS) and cell wall synthesis (mainly cellulose and callose synthesis). In a similar manner, the next step is to study mutants for these respective pathways. Lastly, for BR5, it was noticed that it was able to produce cobalamin, which has previously been linked to drought tolerance, yet the mechanism behind this is still elusive. For all three of these bacterial strains, an untargeted metabolomics experiment will also be performed, which will give us additional insights into their MoA.

We also have two bacterial strains, E7 and E14, that were obtained through collaborations with universities in Egypt and Saudi-Arabia. Here, the idea was to obtain microbes from leaves of desert shrubs that potentially aid in the drought-tolerance of these shrubs. As these strains were specifically derived from leaf material, the aim is to discover whether their drought tolerance-promoting functionality actually stems from being present in the leaf, rather than the root. Hereto, we are currently in the process of fluorescently labelling these bacterial strains. Currently, we have finalized the cloning of the vectors carrying a gene for a fluorescent protein and we are optimizing a transformation protocol that will allow us to bring the prepared vectors inside these bacteria. Then, we can visualise these bacteria in the different plant parts as well as quantify them using qPCR.