

THE RICE *OSMYB4* GENE IMPROVES DROUGHT TOLERANCE IN TRANSGENIC MAIZE

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A major constraint in maize cultivation is the extensive damage caused by water deficit at flowering. The constitutive expression in transgenic Arabidopsis plants of the rice cold-inducible *Osmyb4* gene has been shown to improve cold and drought tolerance, most likely due to the activation of several stress-inducible pathways and to the accumulation of compatible solutes (e.g. glucose, fructose, sucrose, proline). In the present work, we report the effects of the inducible *Osmyb4* expression on drought- and cold stress-tolerance in maize.

Type-II calli from immature embryos of three maize genotypes (Bo21, B73A and GS3) were transformed by the biolistic method with the pCor15aMyb4 plasmid. The putative transgenics were confirmed by PCR for the presence of the *Osmyb4* gene. One selected transgenic event (GS3-E6-1) was crossed with A188, one of the parental lines of GS3. Homozygous transgenic F₄ plants were used to test drought and cold tolerance.

For drought treatments five-week-old potted plants per each genotype (the GS3-E6-1 and GS3 wild-type) were subjected to a progressively increasing water stress. During this treatment, the soil water potential was monitored by weighing the pot. The relative water content (RWC) was determined on leaves of transgenic and wild-type plants at three levels of water potential: (-5KPa, 1000KPa, and -5000KPa). The samples were collected at each level of water potential and analyzed for the *Osmyb4* expression levels and for the content of sugars, amino acids, proline and ABA. For the cold stress treatment, the wild-type and transgenic plants were treated at 4 °C for three and six days and the leaf samples were analyzed for the same traits. We found that *Osmyb4* increased significantly tolerance to water stress but not to cold. Under drought, *Osmyb4* expression affected sugars, amino acids and ABA accumulation in maize leaves. Namely, we observed higher RWC, sugars, proline/aminoacids ratio and a lower ABA content in transgenic plants, suggesting that *Osmyb4* activates some of the pathways involved in the drought-stress response. Although *Osmyb4* seems able to activate stress-responsive pathways in a number of species (Arabidopsis, apple, *Osteospermum*, tomato), its specific effect depends on the genetic background.