

# A novel *Lolium multiflorum*-*Epichloë tembladerae* association: Evaluation of plant growth, seed production and vertical endophyte transmission under drought and waterlogging conditions

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Endophytes of the genus *Epichloë* are key allies for many cool-season grasses, providing them with significant advantages such as increased resistance to herbivores and tolerance to adverse environmental conditions. *Epichloë tembladerae* is a widely distributed endophyte in Argentina, associated with more than 30 grass species. Recently, a strain of *E. tembladerae* was inoculated into *Lolium multiflorum* and formed a compatible association. This study investigated the impact of the novel symbiont *E. tembladerae*-*L. multiflorum* on grass host performance and its stability under waterlogging and drought conditions. Plant growth, biomass, seed production, and vertical transmission of the endophyte to plant progeny were evaluated.

Seeds of *L. multiflorum* of three symbiotic statuses: 1) endophyte-free, 2) colonised with *E. occultans* and 3) colonised with *E. tembladerae* were germinated on filter paper at 15°C. Ninety seedlings per symbiotic status were sown in potting mix and grown at a soil water content of 80% field capacity for four months. All plants were trimmed 5 cm above the crown, and their aerial biomass dried and measured. Thirty plants from each symbiotic status were then subjected to one of three water treatments: 1) drought (40% field capacity), 2) waterlogging (2 cm of water above the soil surface), and 3) control (80% field capacity). After 20 days of treatment all the plants were cut to evaluate regrowth

and half of the plants were harvested to measure root and shoot biomass. The remaining plants were allowed a 15-day resting period before resuming the water stress treatments, which were maintained until seed harvest. At the end of the experiment, seed yield and endophyte transmission frequencies were evaluated. No significant differences in regrowth, shoot and root biomass production, and seed yield among the three symbiotic statuses were detected. However, the drought treatment significantly reduced plant regrowth, biomass production and seed yield compared to the control and waterlogging conditions. Transmission efficiency of *E. occultans* and *E. tembladerae* to seeds exceeded 90% in both cases, and under all water treatments.

These results suggest that water deficiency plays a crucial role in the productivity of *L. multiflorum* and that the association with *Epichloë* has no effect in drought tolerance. Interestingly, the novel association between *L. multiflorum* and *E. tembladerae* resulted in compatible associations even under unfavourable environmental conditions and no negative effects on the plants were observed. These findings expand the knowledge about the specificity between *Epichloë* and their host plants. Considering endophytes in forage and crop breeding, these findings encourage future research to study the effect of *E. tembladerae* on novel grass hosts under different stresses and environmental conditions.