## Evaluation and Development of Poa Germplasm for Salt Tolerance

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## **Objectives:**

- 1. To identify salt-tolerant Poa germplasm that can be incorporated into breeding and genetics efforts.
- 2. To identify genes whose RNA transcript levels vary between control and high-salinity treatments.

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One of the greatest challenges confronting the turf industry is water. Irrigation water is in short supply due to the rapidly growing population, especially in the North American West. Golf course superintendents and other landscape managers are being asked to deal with a very difficult situation--to use less irrigation water, use lower quality water sources, and allow more use of the turfgrass areas. Therefore, turfgrass with high turfgrass quality and greater salt tolerance is essential to meet these expectations.

Extensive breeding efforts in Kentucky bluegrass (Poa pratensis L.) have been conducted by both universities and private companies to improve turfgrass quality traits and pest resistance and some stress tolerance traits. However many Poa species, and most of the available germplasm within Poa pratensis, has not been rigorously evaluated for salt tolerance and turf characteristics in the arid West climate. Similarly, there is not a good understanding of the genetic control of salt tolerance in Poa. Germplasm screening and detailed genetic studies of salinity tolerance are needed for breeding programs to effectively develop salt-tolerant bluegrasses.

We assembled a large and diverse collection of *Poa* germplasm, check varieties, and other species to evaluate in field conditions and controlled-environment salinity evaluations. Seedlings were subjected to increasing salt concentrations until the last plants had been killed by the treatment.

We also conducted suppressivesubtractive hybridization (SSH) to evaluate genes that are differentially expressed under salt stress. We selected two accessions identified in the screening procedure, one identified as salt tolerant and one salt intolerant. We then applied salt stress to



Researchers are identifying salt-tolerant Kentucky bluegrasses that are adaptable to the arid West conditions.

one set of each accession with another set in control conditions-no salt stress.

RNA was extracted from shoot/crown and root tissues, purified, quantified, and then sequenced and analyzed to identify the differentially expressed genes in the tissues. The resulting sequences of the gene transcripts are being compared to a plant sequence database to assign identities to the transcripts based on sequence homology.

Based on two runs of the salinity screening experiment, several germplasm sources were identified with consistently high-salinity tolerance during both runs of the study. The salinity tolerance of some of the germplasm sources was similar to that of the tall fescue and perennial ryegrass check entries. The better performing germplasm has been moved into greenhouse and field hybridization experiments.

The SSH procedure was completed for eight samples making four comparisons of salt and non-salt stressed tissues in both shoot/crown tissues and root tissue. Comparisons were made in "both directions", meaning that genes over-expressed in salt-treated, and over-expressed in control conditions, were investigated. The SSH libraries were cloned, and over 700 clones were selected and sequenced. After comparison to gene databases, the majority of sequences were identified. Most of these annotated genes were involved in some type of plant stress. Some have roles in stress response, some are unidentified, and some only have identities to other stress-induced gene libraries.

The many genes that can be further investigated provide important insights into how Kentucky bluegrass responds to, and tolerates, salt stress at a molecular level. In addition, over 100 sequences had no similar gene found in other grasses. Additional validation of a subset of genes is underway using quantitative real-time PCR (qPCR), wherein we will test differential expression of genes with known salt-response functions.

## **Summary Points**

• We have observed significant variation in salt tolerance among a diverse set of *Poa* species and *Poa pratensis* germplasm.

• The most salt-tolerant accessions exceeded the tolerance of Kentucky bluegrass check varieties and approached or exceeded the tolerance of perennial ryegrass and tall fescue check varieties.

• Over 700 genes related to salt response were cloned and sequenced, some with known involvement in salt tolerance in other plant species.