The experiment was a 4-rep split plot design.

September 2014. Soil type was a sandy golf-course green. Either of these treatments over a 4-week period in with micronutrients included. Plants were irrigated with either of these treatments over a 4-week period in September 2014. Soil type was a sandy golf-course green. The experiment was a 4-rep split plot design.

In this experiment we test if the gene expression of these two genes is higher in tolerant turf species and cultivars vs. susceptible species and cultivars under salt stress.


tables

<table>
<thead>
<tr>
<th>SPECIES</th>
<th>CULTIVAR</th>
<th>SALT TOLERANCE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Poa pratensis (Kentucky bluegrass)</td>
<td>Midnight</td>
<td>Susceptible</td>
</tr>
<tr>
<td></td>
<td>p603</td>
<td>Tolerant</td>
</tr>
<tr>
<td>Lolium perenne (Perennial ryegrass)</td>
<td>Linn</td>
<td>Susceptible</td>
</tr>
<tr>
<td></td>
<td>SR4600</td>
<td>Tolerant</td>
</tr>
<tr>
<td>Puccinilla distans (Alkaligrass)</td>
<td>Fultz</td>
<td>Tolerant (check)</td>
</tr>
</tbody>
</table>

The treatments: 9 dS/m was applied for salt stress, and 1 dS/m for control. Both solutions contained appropriate fertilizer with micronutrients included. Plants were irrigated with either of these treatments over a 4-week period in September 2014. Soil type was a sandy golf-course green. The experiment was a 4-rep split plot design.

After 4 weeks of treatment, plant cores were harvested, quickly washed (<30 seconds), shoot were separated and flash frozen in liquid nitrogen.

RNA was extracted using the Zymo RNA extraction kit.

Reverse transcription used a Fermentas Maxima kit.

qPCR used APEX qPCR master mix with SYBR-green dye.

Beta-tubulin primers were used as controls for relative gene expression.

Relative expression values between salt treated and control samples were then compared.

This is an example of real-time PCR output. The cycle (x-axis) at which the log fluorescence (y-axis) reaches a threshold (dash line) is proportional to the starting amount or gene expression.

The primers for both genes amplified across three species with similar amplification efficiencies.

There were no species differences in gene expression.

The most susceptible sample, Kentucky bluegrass cv. Midnight, showed decreases in expression of both genes under salt stress.

The anti-oxidant gene Thioredoxin was somewhat upregulated in all other samples, consistent with predicted response to salt stress.

1) Expand tests to other cultivars to see if the gene expression profiles are ubiquitous.

2) Test similar genes. There are other vacuolar-type ATPase subunits, and Thioredoxins are also part of a large gene family.

3) Make salt-tolerant x elite hybridizations and begin to improve current elite turfgrass cultivars using marker assisted selection.

References
