

Molecular Characterization of Chinch Bug-Resistant Buffalograsses

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Objectives:

1. Assess the role of oxidative enzymes, specifically peroxidases, in the defense response of buffalograsses resistant to the western chinch bug.
2. Increase the genomic resources available for buffalograss using next generation sequencing technology.
3. Identify genes differentially expressed between chinch bug-infested and noninfested buffalograsses through the use of normalized and subtracted cDNA libraries for susceptible and resistant plants.

Start Date: 2010

Project Duration: three years

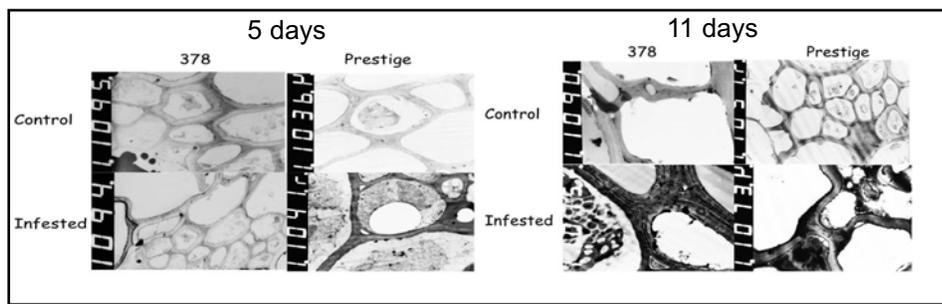
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Turfgrass resistance (specifically tolerance) to insects, when used as part of an integrated pest management program, offers the opportunity to effectively and economically reduce chinch bug infestations while dramatically reducing pesticide inputs. Unfortunately, deployment of chinch bug-resistant turfgrasses has been seriously hampered by insufficient knowledge of plant resistance mechanisms and genes contributing to the resistance.

This information is fundamentally important for formulating plant breeding strategies, and subsequently developing chinch bug-resistant germplasm. In addition, knowledge of specific resistance mechanisms would be valuable for identifying biochemical and physiological markers for use in germplasm enhancement programs, and for characterizing plant defense strategies to insect feeding.

Plants have developed defense strategies to overcome the abiotic and biotic stresses to which they are exposed. One component of this defense system uses a wide array of stress-related proteins which can be elevated or repressed in response to specific or general stress conditions. Changes in the expression of these proteins can play direct or indirect role(s) in the plant's defense response to stress. Reactive oxygen species (ROS), such as hydrogen peroxide, are known to be important early signals for altering gene expression patterns in plant cells.

Despite the benefits gained from molecules like hydrogen peroxide as defense signals, accumulation of these ROS can be toxic to cells. To protect themselves from the effects of ROS accumulation, plants have developed defense-related enzymes (peroxidases and catalases)



Peroxide accumulation at 5 and 11 days after initiation of chinch bug feeding on 'Prestige' and '378' buffalograss.

that break down the ROS.

Previous research by our group has documented increased levels of peroxidases following chinch bug feeding in the resistant (tolerant) buffalograss, 'Prestige', and a loss of catalase activity in the susceptible buffalograss, '378'. These findings support our working hypothesis that a primary plant defense response to chinch bug feeding is to elevate the levels of specific oxidative enzymes, such as peroxidase, to help detoxify peroxides that accumulate as a result of plant stress.

We are documenting the accumulation of peroxides in both resistant ('Prestige') and susceptible ('378') buffalograsses in response to chinch bug feeding. Buffalograss plant tissue was fixed, sectioned, and stained for microscopy. Preliminary results indicate a greater accumulation of peroxides in 'Prestige' infested plants compared to 'Prestige' control plants starting at day 5 and continuing through day 8. Infested '378' plants had similar levels of peroxide accumulation compared to control plants at both day 5 and 8. Both infested '378' and 'Prestige' plants exhibited greater peroxide accumulation compared with their respective control plants at day 11.

These findings suggest that peroxides are playing multiple roles in the plant. Peroxides are likely accumulating in susceptible infested plants in response to the plant's inability to detoxify the ROS. On the other hand, the peroxides in the

resistant infested plants are likely activating plant defense pathways and/or detoxifying ROS.

Research efforts are also underway to increase the genomic resources available for buffalograss using next generation sequencing technology and identify candidate transcripts that may serve as markers for selecting buffalograsses with improved chinch bug resistance. A final objective of this research is to identify specific genes conferring resistance to western chinch bug through the development of cDNA subtractive libraries for resistant and susceptible buffalograsses and gene expression studies.

Summary Points

- This research will (1) allow comparison of gene expression between resistant and susceptible buffalograsses and serve to identify genes differentially expressed in response to chinch bug feeding, (2) provide insights into the biological pathways impacted by chinch bug feeding and help elucidate plant tolerance mechanisms, and (3) facilitate development of improved buffalograsses with tolerance to chinch bugs through marker-assisted selection.

- This research will also shorten the timeframe needed to identify and improve buffalograsses with superior chinch bug resistance, and, because of shared genomics among members of the grass family, may contribute to similar improvements in other grasses species.