## Development of Transgenic Creeping Bentgrass Resistant to the Major Pathogenic Fungi

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## Goals:

- Express our cloned chitinase gene in E. coli, and purify and collect chitinase protein.
- Identify the level of chitinase required to control three major turfgrass pathogens.
- Transform bentgrass with plasmids containing the chitinase gene.
- Evaluate the transgenic plants for resistance to major turfgrass pathogenic fungi.

Dollar spot (Sclerotinia homoeocarpa), brown patch (Rhizoctonia solani) and Pythium blight (Pythium aphanidermatum) are major pathogenic diseases of turfgrass. All of these pathogens contain chitin. The laboratory team has cloned and characterized a full-length chitinase gene that contains the necessary chitin-binding domain ([1; 2; 3] Gene Bank Number L22032). This laboratory also has constructed several plasmids containing a potato proteinase inhibitor controlled by different (wound-inducible and constitutive) promoters. We also have obtained genes for drought resistance and bialaphos resistance from other laboratories.

We have genetically engineered tobacco as a model system, and then creeping bentgrass, with the chitinase gene construct that was developed in our laboratory. We also engineered creeping bentgrass with a few other useful genes including protease inhibitor II, *bar* (bialaphos resistance), and the mannitol dehydrogenase (drought tolerance) genes.

The herbicide resistance of these transgenic plants was confirmed over two years ago. These plants have been transferred to our field facilities, as well as to a seed company's field facilities. We examined the chitinase-transgenic plants containing the chitinase gene for their resistance to brown patch, and bar-transgenic plants after they were sprayed with bialaphos for dollar spot, brown patch, and Pythium diseases. Preliminary work on chitinase-transgenic plants at the greenhouse level indicated

tolerance to brown patch. Furthermore, the results of our inoculation studies have shown that after bialaphos application, bar-transgenic creeping bentgrass have reduced weed problems, dollar spot and brown patch diseases at the greenhouse

level. Eventually, all these transgenic plants must be crossbred and tested for their resistance to the major pathogenic diseases at the field level.

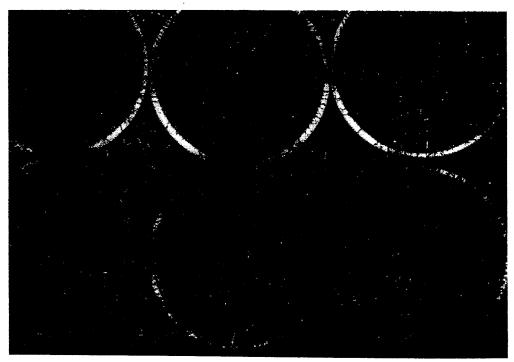


Figure 3. Results of preliminary research on resistance of chitinase-transgenic creeping bentgrass to brown patch (*Rhizoctonia solani*) disease. Upper row: control plants; Lower row: chitinase-transgenic plants.