Bacterial Populations and Diversity withing New USGA Putting Greens

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Goals:

- *Determine bacterial populations associated with putting green root-zone mix materials.*
- *Determine bacterial populations of the rootzone mixes before and after fumigation.*
- *Compare rhizosphere bacterial populations on two different turfgrasses - bentgrass and bermudagrass..*
- *Compare rhizosphere bacterial populations of bentgrass in two different locations - Alabama and South Carolina.*
- *Compare rhizosphere bacterial populations of bermudagrass in two differrent locations - southern Florida and nortthern Florida.*
- *Compare thatch development, rooting and bacterial population of bentgrass in relation to rootzone mix and nitrogen fertilization.*
- *Compare soil and rhizosphere bacterial populations of rootzone mixes containing various clay sources.*
- *Document rhizosphere bacterial population dynamics on bentgrass and bermudagrass over a four-year time period.*

This is a comprehensive, regional project involving three land grant universities in the southeastern United States. The overall objective is to develop baseline data concerning bacterial composition (population and diversity) of new USGA putting greens, both during and after construction.

University of Florida. In order to accomplish the objectives of this project, it was first necessary to ascertain the best sampling and laboratory methods for identifying a bacteria within putting green root zones. At the University of Florida, various diluents and media for each specific group of organisms were evaluated for enumeration of bacteria.

The best overall diluent to use, across all media, was 0.1% glycerol pyrophosphate with 1% glycerol. The following media will be used for enumeration:

a) S-1 medium for fluorescent pseudomonads,

b) selective medium for *Stenotrophomonas maltophilia (*formerly *Xanthomonas maltophilia),*

c) reduced arginine soluble starch medium for actinomycetes,

d) solidified 1/10 strength tryptic soy broth for total bacterial counts,

e) Azide blood base agar for gram-positive bacterial counts,

f) Crystal violet agar for gram-negative bacterial counts, and

g) dilutions heated for 10 minutes at 80 C followed by plating on solidified 1/10 strength tryptic soy broth for heat tolerant bacteria such as *Bacillus* spp.

A protocol was established for all the project cooperators to follow.

Clemson University. Mark Stoddard, CGCS, superintendent of the Charlotte Country Club, has agreed to be a cooperator for this project. The greens were rebuilt to USGA guidelines in the spring and summer of 1996 and seeded with bentgrass in early September 1996. Soil and root samples will be collected from the four bentgrass greens. Assessments of turfgrass quality will be made throughout the experiment.

Auburn University. Miniature greens (1 m by 0.5 m by 1 m), each separately drained to allow collection of leachate, have been constructed. Specific treatments at the Auburn site are two nitrogen rates (1 or 2 lb N/1000 ft^2) and two putting green mixes (100% sand and a 80:20 sand:peat mix). CRENSHAW creeping bentgrass was established on the experimental putting green area.

Soil and root samples will be collected from golf course and university research sites during the months of November, February, May and August. Samples will be subjected to the standardized dilution plating techniques and media developed at University of Florida. Selected rhizobacterial isolates will be identified using GC FAME analysis. Soil and leachate samples will be analyzed for NO_3-N and NH_4-N concentrations using 2M KCI extractions (soil) and standard calorimetric techniques (soil and leachate).