

Analysis of Genetic Diversity in Rough bluegrass, Colonial and Velvet Bentgrasses Using RAPD Markers

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Introduction

Rough bluegrass (*Poa trivialis*) is a cool-season perennial grass that is grown in wet shaded locations for home lawns, golf courses and sports fields. It is also used for winter overseeding of warm season turfgrasses in south where it forms one of the highest quality turf of any cool-season turfgrasses. Colonial bentgrass (*Agrostis capillaris*) is a sod-forming, cool-season perennial grass, primarily grown in northeastern and northwestern regions of the United States. This fine textured grass spreads by short rhizomes and stolons to form close tight turf and is best suited to golf course fairways. Velvet bentgrass (*Agrostis canina*), the finest textured of all bentgrasses, is adapted to cool, moist areas of coastal regions of the United States. It is primarily used on putting greens, where it forms an attractive, low-growing, compact turf with a high-quality putting surface. It is also the most shade-tolerant of all the bentgrasses and is used on fairways where nitrogen levels are kept low. All of these three species are not extensively utilized, partly due to the lack of breeding efforts. Germplasm of turfgrass species, including the aforementioned three species, are being collected by USDA plant introduction stations throughout the world. However, these germplasm are largely uncharacterized genetically. To better utilize these collections, genetically characterizing each accession of these species is critically important. Molecular markers are a powerful, reliable and cost effective tool for evaluating genetic diversity and can provide important insights for breeding. The objectives of this research were to: (1) evaluate the genetic diversity of selected accessions and varieties of rough bluegrass, colonial bentgrass and velvet bentgrass using RAPD (Randomly Amplified Polymorphic DNA) markers, and (2) evaluate the morphological variations of these materials under field conditions.

Materials and methods

This research involves 61 accessions, including 27 accessions of rough bluegrass, 27 accessions of colonial bentgrass, and 7 accessions of velvet bentgrass that were obtained from the Western Regional Plant Introduction Station (USDA) at Pullman, WA. All seeds were sown in cell packs and DNA was extracted from young leaves after growing one-month in the greenhouse. After the seedlings reached a desirable height, they were transplanted to the Iowa State Horticulture Research Farm for further studies on morphological characteristics under field conditions. All accessions were planted in a randomized complete block design with a maximum of 18 replications for each accession.

RAPD analysis is based on the polymerase chain reaction (PCR). After DNA extraction, PCR conditions were optimized for RAPD primers by examining at least four accessions. Eighty primers were screened for polymorphism. Fifteen out of 80 primers that showed high polymorphism were used to analyze the 27 accessions of rough bluegrass.

The PCR products were checked by agarose gel electrophoresis. The polymorphic bands were scored either as “1” for presence and “0” for absence and a similarity coefficient matrix was formed. This matrix was analyzed by the “Mesquite” software to construct dendrogram and principal coordinate analysis. Using this similarity coefficient matrix, genetic distance was analyzed by the “PAUP” software.

Results

Dendrogram analysis showed that accessions in three out of five clusters in the dendrogram tree have a positive relationship with their geographic origins. This is confirmed by their genetic distances. Genetic distances showed that accessions from the United States and Iraq have high genetic distance between them. Accessions from Denmark and Yugoslavia have low genetic distance between them. Data on morphological characteristics of these accessions is unavailable at present.

For colonial and velvet bentgrasses, DNA was extracted and PCR was optimized. Primers were screened and 18 out of 80 primers that were screened showed high polymorphism. Data on genetic distance and morphological characteristics of these accessions is unavailable at present.