Application of Molecular Diversity in a Forage Grass Breeding Program

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Introduction Little or no genotypic information is available for many forage grass populations. The degree of genetic similarity within and among populations greatly influences the choice of breeding strategies and germplasm for developing improved cultivars. Molecular markers have proven effective in classifying genetic diversity of a number of perennial grasses (e.g. Fu *et al*., 2004; Kubik *et al*., 2001). We present here an overview of our efforts to integrate molecular diversity data into our breeding program.

Materials and methods Amplified fragment length polymorphism (AFLP) markers were used to examine genetic variation among tall fescue (*Festuca arundinacea*) populations (Mian *et al*., 2002), as well as among and within hardinggrass (*Phalaris aquatica*) populations (Zwonitzer *et al*., 2003). Expressed sequence tag – simple sequence repeat (EST-SSR) markers originating from tall fescue were used to classify genetic variation among and within populations of Canada wildrye (*Elymus canadensis*) and Virginia wildrye (*Elymus virginicus*) (Saha *et al*., 2004). Standard statistical analyses, such as Nei and Li (1979), were used to classify within and among population genetic variation.

Results Tall fescue populations from the southern Great Plains were found to differ genetically from several cultivars including KY-31 (Mian *et al*., 2002), leading us to make further collections of tall fescue germplasm from this region. The pattern of genetic variation between hardinggrass populations agreed closely with geographical origins. We have used this information to construct a population based on accessions from Morocco, and a broad population based on accessions from a wide geographical area. We intend to determine in the future if these two populations represent potential heterotic groups. Genetic variation was minimal within wildrye populations, indicating that a given wildrye population can be handled as a pure line in our breeding program. Canada wildrye populations heavily infected with an endophytic fungus clustered together; further investigation revealed that presence of the endophyte did not bias interpretation of the plant genetic variation data. Sequencing of a specific locus indicated that single nucleotide polymorphisms (SNPs) were the source of variation among wildrye accessions.

Conclusions Genetic diversity data has proven to be valuable to our breeding program in directing acquisition of germplasm, constructing populations, and choice of breeding methods. In the future marker diversity data may also aid in identifying heterotic groups in forage grass species. The application of high throughput marker systems, such as SSR's developed from tall fescue, and the development of additional markers such as SNP's, will continue to enhance our ability to understand and utilize genotypic diversity in the forage grasses.

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