The development of marker assisted selection breeding strategies in *Lolium perenne* L.

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Abstract

The successful incorporation of maker-assisted selection (MAS) into current breeding strategies for outcrossing species, demands a reduction in the number of parents used to develop a synthetic cultivar and a greater understanding of how to utilise the genetic diversity present in such species.

To optimise parent number used in synthetic cultivar development, eight parents of a perennial ryegrass (*Lolium perenne* L.) synthetic cultivar were chosen to determine parental performance contribution for the cultivar. Parents were evaluated for their degree of genetic similarity/dissimilarity using amplified fragment length polymorphism (AFLP) analysis. Three genetic groupings were observed. This information has been used to re-construct the effect of parental combinations on the synthetics performance based on parental similarity/dissimilarity.

Keywords

amplified fragment length polymorphism (AFLP)

Introduction

Perennial ryegrass is a wind-pollinated, self-incompatible species. Cultivars are developed from the random mating of a number of elite individual plants (1). However, such a breeding strategy is complex and not directly suited to the implementation of molecular breeding strategies.

Forage plant breeders tend to select large numbers of parents to avoid inbreeding depression and to maintain the diversity required for the cultivar to be broadly adapted to a wide range of different environments (2). However, successful lucerne (*Medicago sativa* L.) cultivars may be derived from small numbers of parents (3).

This study is developing strategies to allow breeders to use DNA-profiling techniques for the determination of parental contribution to the performance of a new cultivar and thereby enable breeders to reduce the parental numbers used for cultivar development.

Materials and methods

The synthetic cultivar of perennial ryegrass, KT(2), was chosen due to its broad adaptation to a number of Australian environments and access was available to the original eight parents (4).

An 8 x 8 partial diallel pair crossing system, without selfing (self-incompatible species), was used to produce a total of 28 crosses.

It was not practical to re-construct all of the potential synthetic parental combinations due to the large number of possible combinations based on synthetics from between three and eight parents. Therefore, synthetics based on the five most similar and dissimilar parental combinations for each parental number were constructed. Each synthetic combination was developed by combining an equal number of seeds from the pair crosses involved.

AFLP profiling analysis was conducted on these genotypes to determine the degree of genetic similarity/dissimilarity between the parents.

Each synthetic population is currently being grown in the field as a 64-plant microsward (as described by Foster (5) with some modifications). The original KT(2) cultivar has been included in this study for comparisons with the synthetic combinations. Herbage dry matter yield will be measured over a two year period.

Results and discussion

AFLP profiles were generated for the nine KT(2) parental genotypes. Analysis of AFLP profiles of ten primer combinations identified 760 polymorphic bands that were scored. The profiles of another six primer combinations were not analysed due to the detection of a large number of poorly resolved fragments. The coefficients of similarity among parental genotypes ranged from 0.53-0.68 based on the average taxonomic distance (Figure 1). The phenogram and principal coordinates analysis (Figure 2) showed separation of the parents into three major clusters. Group 1 consists of parents 1 and 12; Group 2 consists of parents 5, 17, 18 and 26; while Group 3 consists of parents 2 and 30.

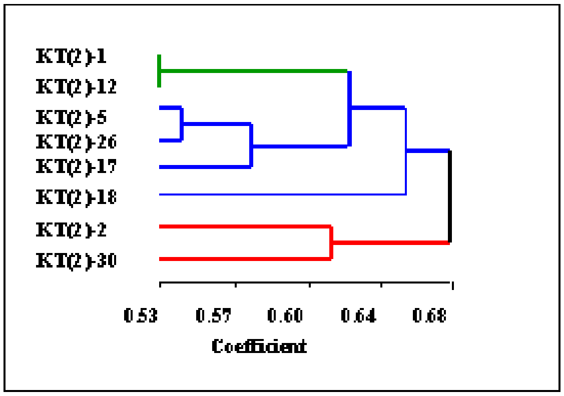

Figure 1: Phenogram of KT(2) parents based on AFLP variation.

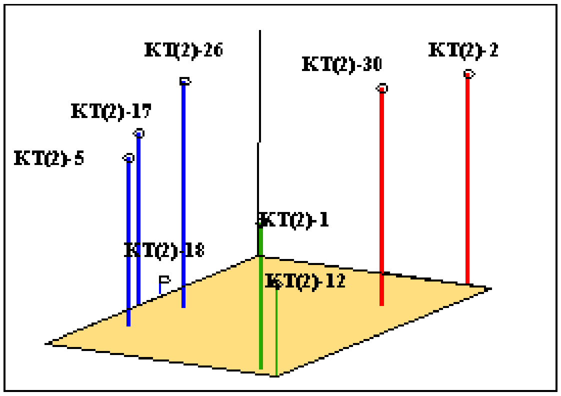

Figure 2: Principal coordinates analysis based on the AFLP variation of the KT(2) parents.

Based on these three clusters, parental combinations have been constructed according to genetic similarity or dissimilarity. In lucerne, Hill and Elgin (3) were only able to indicate a range (4 to 16) for the optimum number of parents per synthetic as they did not test all of the parental numbers. This study used a synthetic cultivar developed from a smaller number of parents, with all parental numbers being tested.

A number of general breeding strategies are available, that use MAS to select for target genes within the polycross progeny of a small number of parents (2). Therefore, this research will allow breeders to use DNA profiling techniques for the determination of parental contribution to the performance of a new cultivar and thereby enable the reduction of parent numbers used for MAS during variety development.

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