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Does AFLP diversity reflect consanguinity within meadow fescue breeding material?

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Introduction Cultivars of perennial grass species are usually synthetics with a limited number of constituent parental clones, prone to inbreeding depression. Plant breeders aim at a balance between intensity of selection and maintenance of genetic diversity when making their choice of parent clones in an intuitive way, aided by fragmentary pedigree information. Molecular markers offer new opportunities for assessing genetic diversity among selected plants. The objective of the investigation presented here is to check if the genetic distance as measured by AFLP polymorphisms reflects consanguinity among *Festuca pratensis* individuals from our breeding programme.

Materials and methods 255 genotypes of *Festuca pratensis* originating from seven, partly interrelated gene pools (Table 1) were analyzed. AFLP templates were prepared by restriction digestion (*Eco*RI and *MseI*) and adaptor ligation of 1 μ g genomic DNA. For selective amplification, 11 combinations of 6 EcoRI and 2 MseI primers with 3 additional nucleotides each were used. Visual scoring resulted in 272 polymorphic markers. Genetic diversity among genotypes was calculated using Euclidean squared distances. The SAS v. 8.02 package was used for further statistical analysis.

Table 1 Origin of plant genotypes and mean Euclidean squared distances (E²) within gene pools

Gene pool	no.	Programme	Origin	E^2
Ecotypes 99/03 S1a S1b S2 S3 cv. Preval	31 59 46 62 11 24	FAL Schmidt Schmidt Schmidt Schmidt Badoux	Collection in 28 different permanent meadows F1 progeny from random mating of 70 plants F1 progeny from random mating of 65 plants F1 progeny from random mating of 60 plants various plants from recurrent selection schemes Syn4 progeny from polycross with 16 clones	76.3 65.9 66.6 65.4 66.2 62.0
cv. Pradel	22	Badoux	Syn4 progeny from polycross with 15 clones	58.2

Results AFLP marker diversity within gene pools reflected the intensity of selection (Table 1). Non-related ecotypes showed the greatest diversity, followed by random mating breeding populations and narrow-based cultivars. Principal component (data not shown) and cluster analysis of AFLP data (Figure 1) clearly separated the gene pools and reflected known pedigree relationships among them. Half-sibs and half-cousins were significantly less distant to each other than to comparable non-related plants (Table 2). Average distances between half-sibs were 86.9 % and between half-cousins 93.5 % of the distances observed between pairs of unrelated plants. This corresponded well with the expected co-ancestry coefficients of 12.5 % for half-sibs and 6.25 % for half-cousins.

Table 2 Euclidean squared distances (E^2) between pairs of consanguineous and non-related plants. E^2 values within one row not followed by the same letter are significantly different.

Gene	half-sibs		half-cousins		non-related
pool	no.	E^2	no.	E^2	E^2
S1a+S1b	31	58.1c	44	64.4b	67.4a
S2	17	60.1b	21	61.0b	68.1a
both	48	58.8c	65	63.3b	67.7a

Figure 1 Relationship among gene pools as revealed by cluster analysis of AFLP data



Conclusions There were two distinct lines of evidence suggesting that AFLP diversity reflects the degree of consanguinity within *Festuca pratensis* breeding material: grouping into gene pools by cluster analysis, and reduced distance between pairs of consanguineous plants. AFLP analysis can therefore be used to optimize genetic diversity among parent clones of synthetic cultivars.