

Improving the utilisation of germplasm of *Trifolium spumosum* L. by the development of a core collection using ecogeographical and molecular techniques

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Introduction A core collection is a sub-set encompassing more than 70% of the variability of all accessions held in a collection (Brown 1995), the development of one for *Trifolium spumosum* (bladder clover) could assist in future development of the cultivar within southern Australia. The aim of this work is to develop a core collection of *Trifolium spumosum* as a model for other pasture legume species using molecular and ecogeographical data.

Materials and methods Accessions with near complete ecogeographical data were selected from the Australian *ex situ* collection of *Trifolium spumosum*. This collection of 317 accessions was grouped into 5 geographical regions. MStrat Software (Gouesnard *et al.*, 2001) was used to select the preliminary core of 30% of the collection. Fluorescent Amplified Fragment length polymorphism (FAFLP) will be used to screen the diversity within the species. The primers producing the highest number of bands will be used to screen the preliminary core collection. Mstrat will be used to develop final core collections containing 30% of the preliminary core.

Results A preliminary core collection of 95 accessions was selected. In the randomly selected cores the scores (based on Nei index) were different for each repeat, however, scores were constant for cores selected using the maximising strategy (OPT in Table 1). A final core of 32 accessions will be selected using AFLP and ecogeographical data. The AFLP markers with the green fluorescent labelled EcoR-I primer (TET) showed the greatest amount of data with the highest diversity (Figure 1). The genetic profiles of the preliminary core will be scored and recorded in a database with ecogeographical data.

Table 1 Active scores generated from the optimisation (OPT) and random (RAN) sampling methods using MStrat for core sizes of 76 and 109

Core size	Method	Final score [#]								
		1	2	3	4	5	6	7	8	9
76	OPT	101	101	101	101	101	101	101	101	101
	RAN	84	85	84	83	82	84	83	80	81
109	OPT	101	101	101	101	101	101	101	101	101
	RAN	89	91	92	82	88	90	91	92	86

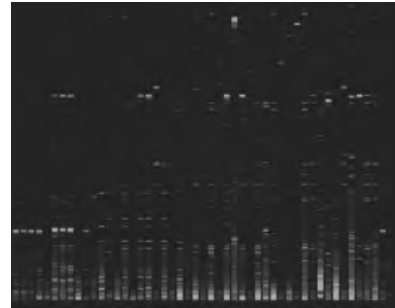


Figure 1 AFLP bands obtained from 48 samples in green fluorescent labelled EcoRI primers

Conclusions The present study hopes to demonstrate that a combination of AFLP marker and ecogeographical data can be used to develop an effective core collection that maintains the majority of the genetic diversity. This model should be used to develop core collections of other pasture legume species that are too large for efficient utilisation.

References

- Brown, A.H.D. (1995). The core collection at the crossroads. In: T. Hodgkin, A.H.D. Brown, Th. J.L. van Hintum, & E.A.V. Morales (eds.) Core collections of plant genetic resources John Wiley & Sons, 3-20.
- Gouesnard, B., T.M. Bataillon, G. Decoux, C. Rozale, D.J. Schoen & J.L. David (2001). MSTRAT: an algorithm for building germplasm core collections by maximising allelic or phenotypic richness. *Journal of Heredity*, 92, 93-94.