Enhanced drought tolerance in white clover

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Abstract

Drought-stress limits white clover (*Trifolium repens* L.) persistence in many New Zealand regions. As a component of breeding for enhanced drought tolerance, 8 selection groups (110 lines in total) of white clover were evaluated in the Wairarapa over a 2-year period. The selection groups included Australian white clover ecotypes, selections from New Zealand dryland populations, root morphology selections, pre-release selections from New Zealand breeding programmes, and existing overseas and New Zealand cultivars. The selection groups derived from New Zealand dryland populations had the highest forage yield and plant survival, 21 of the 24 individual lines with >30% plant survival coming from these groups. Groups containing Australian ecotypes and overseas cultivars had the lowest forage yield and plant survival. Selections for root morphology *per se* were lower yielding and less persistent than selections made from New Zealand dryland populations evaluated in drought-prone environments. However, some improvements in forage yield and persistence were observed through selecting for root morphology after screening the same New Zealand dryland populations in a drought-prone North Canterbury site.

Keywords: drought tolerance, plant breeding, root morphology, *Trifolium repens*

Introduction

Drought stress limits white clover (*Trifolium repens* L.) persistence in many New Zealand regions. In regions with severe moisture stress annual legumes such as subterranean clover are more suitable than white clover (Macfarlane *et al.* 1990b; Sheath et *al.* 1990); however, in areas where droughts are less severe, a more persistent white clover cultivar is needed. Identification of white clovers with enhanced drought tolerance could increase legume content in grazed swards, enhance pasture quality and animal performance. A wide range of white clover germplasm has been screened for drought tolerance in New Zealand, including more than 100 white clover populations evaluated on southern Hawkes Bay and North Canterbury farms (Woodfield & Caradus 1987; Woodfield, unpublished data). Populations collected from drought-prone New Zealand farms had higher persistence and productivity than existing New Zealand and overseas cultivars (Woodfield & Caradus 1987). These dryland populations were also more tap-rooted than was expected from their shoot morphology (Caradus & Woodfield 1986). In a national evaluation of 10 white clover lines, lines that maintained high stolon densities generally performed best; however, no line survived at two severely stressed sites, and line performance was inconsistent at moderately stressed sites (Chapman et *al.* 1986). Reseeding ability was an advantage in the driest environments (Macfarlane *et al.* 1990b) but benefits were less evident in moderately stressed environments (Chapman & Williams 1990; Widdup & Turner 1990). Plant recovery from buried stolons (Chapman & Williams 1990) and the production of large nodal roots (Macfarlane *et al.* 1990a) were other traits implicated in drought tolerance, but the relative importance of these factors has yet to be determined.

The objectives of the current study were to evaluate the persistence and productivity in a drought-prone Wairarapa environment of: (i) selections from New Zealand dryland populations, (ii) root morphology selections, (iii) ecotypes collected from drought-prone regions of Eastern Australia and (iv) cultivars and pre-release cultivars with potential drought tolerance.

Materials and methods

Plant material

110 white clover lines, comprising 8 groups based on germplasm source and the selection criteria, were evaluated. Group 1 consisted of 13 Australian ecotypes which had performed strongly in an earlier trial at Palmerston North. Groups 2-4 contained divergent root morphology, including selections: small or large tap-root diameter, few or many large nodal roots, and low, medium or high root to shoot ratio. Group 2 contained F, progeny of plants selected from a large collection of cultivars and ecotypes (Caradus & Woodfield 1986), and Group 3 consisted of F_2 progeny after a second cycle of selection for root morphology (Woodfield & Caradus 1990). Genotypes that survived vegetatively at a harsh North Canterbury location were screened for root morphology and crossed in isolation to produce the Group 4 lines (Woodfield & Caradus 1987). Groups 5 and 6 were progenies from New Zealand dryland populations,

selected for persistence and productivity in either North Canterbury (Group 5) *or* southern Hawkes Bay (Group 6). The genotypes that were crossed to produce Group 4 lines were a subset of those used to produce the Group 5 lines. Group 7 consisted of 4 overseas cultivars (Crau, Irrigation, Pathfinder, and Zapican) and 2 ecotypes, one from Syria and the other from Gansu Province, China. Seven elite selections from other New Zealand breeding programmes comprised Group 8. All lines in groups 7 and 8 performed strongly under rotational grazing at Palmerston North during a 4-year period (Caradus *et al.* 1991). The number of lines in each group is listed in Table 1. Four New Zealand cultivars (Prop, Grasslands Demand, Grasslands Tahora, and Grasslands Huia) were also included as controls.

Trial site

The trial was located at Te Ore Ore, 5 km north-east of Masterton, on a north-west-facing hillside with a 15^0 slope. The resident sward was perennial ryegrass (*Lolium perenne* L.) dominant, although other grasses, including browntop (*Agrostis tenuis* Sibth.), *Bromus mollis* L., barley grass (*Hordeum murinum* L.), and crested dogstail (*Cynosurus cristatus* L.), were also present. Resident clovers were eliminated from the trial site in the 3 months before establishment by spraying with dicamba. MCPB was also sprayed to eliminate thistles. To reduce. the initial grass competition, glyphosate was used 6 weeks before planting to spray 100-mm wide strips in the resident pasture. Twelve-week-old white clover seedlings were transplanted into these strips on 26 July 1988. Individual plots were 2-m long rows, spaced at l-m intervals, and contained 10 seedlings planted at 200-mm spacings. A randomised complete block design with 5 replicates was used. The site was fenced to preclude grazing during the first 3 months, and was rotationally grazed thereafter with both cattle and sheep. A soil test in June 1988 resulted in application of 180 kg/ha of 30% potassic superphosphate, 25 kg/ha of sulphur, and 2300 kg/ha of lime.

Plant measurements included visual scores (0 = no growth to 5 = highest yielding) of seasonal growth during the first 2 years of the trial, number of plants surviving vegetatively, and spread of surviving plants after 2 years. As plant survival over the whole trial was less than 20% at the end of 2 years, further growth notes were not taken. Analyses of variance were performed on all measurements using SAS procedure GLM for unbalanced data. Forage yield and plant spread were calculated relative to the performance of the Huia control line, and plant survival was calculated as a percentage of the plants that had established 4-months after transplanting. Paired group means were compared using LSDs calculated based on unequal number of lines at P=0.05.

Table 1 Forage yield, plant spread and survival after two years, of eight selection groups and four control cultivars. Forage yield and plant spread are reported relative to the performance of Grasslands Huia.

Selection group	Description	No. of lines	Forage yield (% of Huia)				Spread (% of Huia)		Survival (%)	
			Year 1	Year 2	Total	Range	Mean	Range	Mean	Range
1	Australian ecotypes	13	68 d†	62 b	65 f	17-95	69 ef	O-126	7 e	O-28
2	Rmt-morphology selections (F_1)	9	83 c	107 de	93 d	73-133	111 bc	56169	13 d	4-32
- 3	Root-morphology selections (F_2)	15	81 c	84 g	82 e	19-122	81 e	o-145	13 d	042
4	North Canterbury root-morphology selections	22	101 a	118 be	108 b	65-145	110 be	67-157	29 a	6-72
5	North Canterbury elite selections	10	101 a	112 cd	106 bc	71-126	87 de	64118	29a	10-42
6	Southern Hawker Bay elite selections	24	96 ab	123 b	107 bc	70-139	121 b	74-146	25 ab	6-50
7	Overseas cultivars	6	6Qd	68 h	69 f	51-98	61 f	o-131	5 e	o-9
8	New Zealand pre-release selections	7	100 a	88 g	95 d	81-107	86 de	46-156	15 cd	8-24
	Controls									
	Grasslands Demand		102 a	137 a	117 a		156 a		18 c	
	Prop		92 b	136 a	111 ab		159 a		15 cd	
	Grasslands Tabora		76 cd	91 fg	83 e		88 de		12 d	
	Grasslands Huia		100 a	100 ef	100 cd		100 cd		24 b	

† Means, within columns, followed by the same letter are not significantly different at 0.05 probability level.

Results and Discussion

Of the initial 5500 genotypes established at this site only 19% persisted vegetatively after 2 years. Drought stress was not the only factor that influenced plant performance; the best lines also tolerated the accumulated effects of grazing management, soil fertility, interspecific competition, pests and diseases.

Performance of selection groups

Three selection groups persisted as well as or better than Huia, the most persistent New Zealand cultivar (Table 1). These groups were all progenies of selected genotypes from earlier evaluations at drought-prone North Canterbury or southern Hawkes Bay sites. Their strong performance confirms the previously reported potential of New Zealand dryland populations as a source of drought-tolerance (Woodfield & Caradus 1987). Of the 24 lines with 30% or higher survival rate after 2 years, 21 came directly from New Zealand dryland populations (Groups 4, 5 and 6). Two of the three remaining lines were F, root-morphology selections and the last was an F_2 root-morphology selection (Table 1). The same three groups also had the highest forage yields, being equal to Huia in the first year and significantly higher yielding than Huia in the second year. Five selection groups contained individual lines which yielded as well as or better than the best currently available New Zealand cultivar, lines from Groups 4, 5 and 6 again predominating.

Of the two dryland sites used in the initial cycle of selection, the North Canterbury site was much drier and harsher. Despite this, elite selections from North Canterbury (Group 5) and southern Hawkes Bay (Group 6) performed similarly in the current trial (Table 1). Thus the populations used in the breeding programme may be more important than the severity of the environment in which it is tested. Alternatively, using harsher evaluation sites should decrease the time required to complete each cycle of selection. Regular drought stress increases the selection pressure imposed on these populations.

Selecting drought-tolerant genotypes in drought-prone environments proved more effective than selecting for root morphology *per* se. The F, and F_2 root morphology selections (Groups 2 and 3) had not been screened in dryland conditions, and were less persistent and lower yielding than Groups 4, 5 and 6 which had been screened in either North Canterbury or southern Hawkes Bay (Table 1). The F, root morphology selections were lower yielding than Huia in the first year but improved relative to Huia in year 2 (Table 1). Surviving plants from the F_2 root morphology group had lower yield and spread than the F, selection group (Table 1), probably due to inbreeding depression associated with the small population size used to generate the F_2 selections. The screening method for determining root morphological traits is labour intensive, limiting the number of genotypes which could be screened and used to develop the various selections (Caradus & Woodtield 1990; Woodtield & Caradus 1990).

Mean plant spread was significantly greater for Group 4 than 5, but mean yield and survival were not improved by selecting for root morphology after pre-screening New Zealand dryland populations in North Canterbury (Table 1). However, 3 North Canterbury root morphology selections (Group 4) had better survival, and 4 selections had higher forage yield, than the best North Canterbury elite selection (Group 5), despite the same genotypes being used to develop both selection groups. Incorporating selection for root morphological traits associated with higher yield and persistence should improve white clover performance, provided adequate population size is maintained.

Overseas cultivars and ecotypes

Studies of purported drought-tolerant overseas cultivars such as Clarence Valley, Haifa, Tamar and Louisiana S 1 have shown they are poorly adapted to New Zealand conditions (Chapman et al. 1986; Macfarlane et al. 1990a). Most of these cultivars are large leaved and lack the necessary stolon densities to persist under intensive grazing (Chapman et *al.* 1986). The current group of overseas cultivars and Australian ecotypes was evaluated because they maintained high stolon densities in grazing trials at Palmerston North (Caradus et *al.* 1991; Woodfield, unpublished data). Despite this, both groups had very poor survival, only 2 lines (both Australian ecotypes) exceeding 10% survival. Huia consistently outyielded these groups, the best Australian ecotype yielding 95% of Huia and the best overseas cultivar (Pathfinder) yielding 98% of Huia (Table 1). The mean spread of surviving genotypes for both groups was significantly less than that of Huia, Demand and Prop. None of the cultivars or ecotypes evaluated exhibited any potential adaptation to NZ dryland conditions.

Performance of New Zealand cultivars and pre-releaseselections

Two recently released New Zealand white clover cultivars, Demand and Prop, exhibit some potential for drought-prone environments. Demand and Prop were more productive and had significantly greater spread than Huia in the second year (Table 1). Prop, a small-leaved, early-flowering cultivar with high stolon densities and strong reseeding ability was released specifically for North Island dry hill country

(G. Sheath pers. comm.). Demand, a medium-leaved cultivar with medium to high stolon density, was bred in Southland for improved early-spring and summer performance (Widdup et al. 1989). Despite increased yield and spread, the survival of both cultivars was lower than that of Huia, and significantly lower than that of Groups 4-6 (Table 1). Tahora had the lowest forage yield and persistence of the four control cultivars. Forage yield of the best Groups 2, 4, 5 and 6 lines exceeded those for the best control cultivars, Demand and Prop.

None of the pre-release selections was an improvement on the existing cultivars. The best pre-release selection yielded 7% higher than Huia and had 22% survival, while another pre-release line had slightly higher survival (24%) but poorer overall forage yield. Both of these pre-release selections were bred for high stolon densities in moister environments (Caradus *et* al. 199 1). The poorer performance of these lines suggests that while maintenance of high stolon and node densities is important in drought-prone environments (Chapman *et* al. 1986; Charlton *et al.* 1989) other factors are also important.

Conclusion

Lines with higher yield and survival than Huia were predominantly selections made in drought-prone environments. Selection for root morphology *per se* was less effective than screening in drought-prone environments, although some improvements were observed from selecting for root morphology after screening lines in North Canterbury.

ACKNOWLEDGEMENTS

We wish to thank Chester Meredith and his farm manager Brian McKeown for use of the Lochliddy farm trial site; and Syd Easton, John Ford, Roger Claydon, Tania Wiki and the late Gordon Hardy for assistance in planting this trial.

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