

RESPONSE TO DROUGHT OF WHITE CLOVER LINES SELECTED FOR DIFFERENT STOLON MORPHOLOGIES

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ABSTRACT

White clover (*Trifolium repens* L.) lines were selected from within large and small-leaved cultivars of Grasslands Kopu and Grasslands Tahora, respectively, for long or short internodes, and for high or low branching frequency from plants grown in sun and shade (50% full sunlight). Lines were compared for drought tolerance in a perennial ryegrass (*Lolium perenne* L.) sward in boxes. Prior to imposing drought branching frequency selections did not differ in branching frequency, although the low branching frequency selection had a higher percentage of rooted nodes. After an imposed drought treatment sun-selected lines grew better than shade-selected lines relative to their non-stressed controls suggesting that white clover selected under full-sun may be more drought-tolerant than lines selected in shade. Selections for different stolon morphologies did not differ in stolon and root growth at the end of the drought.

KEYWORDS

Breeding, drought, morphology, selection, stolon, white clover (*Trifolium repens* L.)

INTRODUCTION

White clover (*Trifolium repens* L.) persistence may be improved by selection for high stolon growth rate and/or high node branching frequency (Beinhart et al., 1963). Recent selections for divergent stolon morphology in large-leaved (Grasslands Kopu) and small-leaved white clover cultivars (Grasslands Tahora) offer the potential to evaluate the effect of stolon morphology on moisture stress tolerance. Caradus and Chapman (1991) described selections for extremes of internode length (long and short) and stolon branching frequencies (high and low). Lines were selected either under full sun or shade (50% sun), to provide 16 different lines. The light selection regime is of interest since light influences both stolon and branch development rate. Stolon elongation rate increase and branching frequency decreased when stolons were shaded and/or exposed to a low red to far-red light ratio (Mouliia et al., 1989).

Utilising these different stolon morphological selections the aim was to determine :

- whether differences in stolon morphology of plants selected in monoculture are also apparent when the plants are grown in a perennial ryegrass sward, and
- whether stolon morphology can influence drought tolerance of white clover.

METHODS

Six genotypes per line, chosen to represent each of the 16 stolon morphology selections, were vegetatively propagated using rooted stolon tips. One clone of each of the six white clover genotypes from each line was planted in separate boxes containing 8-week old endophyte-free 'Nui' perennial ryegrass (*Lolium perenne* L.) swards located in a glasshouse. Each box (0.3 m by 0.2 m by 0.2 m) was lined with polythene film. Boxes were filled with a freely-draining Terrace Medifibrsit, a Pongakawa peaty loam of volcanic origin. The soil was weighed and packed in the boxes to a bulk density of 0.75 Mg/cm³ and the equivalent of 60 kg/ha P, 30 kg/ha K, 150 kg/ha S, was added to each box. Time Domain Reflectometer (TDR) rods were inserted into the boxes to monitor the volumetric soil moisture content. After trimming the sward and allowing the plants to establish for one month, water was added to bring the boxes to 59% moisture content using the TDR measurements. In each box the youngest node on one stolon of three plants was marked with an oil-based paint. Boxes were watered every 2 to 3 days and maintained as close as possible to the same moisture content, and herbage was

harvested every 2 to 3 weeks. Marked stolons were harvested after 39 days and the stolon length, number of branches, number of nodes, number of rooted nodes, and clover dry weights were measured. From these measurements, internode length, stolon elongation rate, and percentage of nodes with branches or a root were calculated.

Since the selections across different cultivars differed in size and because we were interested in comparing the growth of droughted plants relative to their paired, non-stressed controls, the absolute plant measures were not compared. Rather, an index of each droughted plant relative to its paired, watered plant was calculated. Some of the watered control plants did not form branches during the drought period, and many of the treatment plants also did not have branches. Because the data were not normally distributed, the mean population of nodes branching on the droughted plants is not reported. Pre-planned contrasts of selection groups were analysed using GLM of SAS. The pre-planned comparisons were: (a) Kopu vs. Tahora, (b) long internode vs. short internode selection, (c) high branching frequency vs. low branching frequency, and (d) sun-selected vs. shade-selected lines. Contrasts (b) through (d) were also made within Kopu and Tahora.

RESULTS AND DISCUSSION

Pre-drought: Prior to imposing the drought treatment, the total herbage (grass and clover) from the eight harvests did not differ for any of the cultivars or pre-planned comparisons. When clover weight was determined separately for harvests 2, 4, and 8, only the Kopu vs Tahora contrast was significant ($p < 0.05$), with Kopu producing 56% more herbage than Tahora. Before the drought, Kopu had significantly ($p < 0.05$) shorter internode length (37 vs. 42 mm), slower stolon elongation rate (5.4 vs. 7.3 mm/day), and a lower percentage of rooted nodes (41 vs. 47%) than Tahora. Internode length (55 vs. 29 mm) and stolon elongation rate (9.1 vs. 4.7 mm day⁻¹) were significantly higher in the long than the short internode selection when compared across cultivars. The percentage of branching nodes and the number of branches did not differ between selections either across or within either of the cultivars. The low branching frequency selection had a significantly ($p < 0.05$) higher percentage of rooted nodes than the high branching frequency selection (45 vs. 35%). Although a difference in branching frequency was not evident during this 5 weeks of stolon growth, it may become evident under different environmental conditions, since branching differences are sensitive to light quantity and quality (Mouliia et al., 1989).

Drought: The sun-selected lines, when compared across cultivars, had higher drought indexes for internode length, stolon elongation rate, and total clover dry weight than the shade selected lines (Table 1). The swards in droughted boxes wilted and collapsed, which conceivably allowed more light or a different red/ for red ratio of light to reach the stolons than in the watered, more upright swards. The longer internodes and higher elongation rates of lines selected in the sun may be due to these plants being more responsive to light than those selected in the shade. The selection for stolon morphologies under different light regimes may produce lines which respond differently to a range of light/shade environments. However, the drought indices of the stolon morphology selection groups did not differ for any of the plant characters measured across or within cultivars (Table 1).

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Table 1

The mean drought index of stolon and root characters, and white clover dry weight between cultivars and across cultivars for each selection group, with the significance of pre-planned contrasts. The drought index is the performance of each droughted plant divided by the performance of its paired, watered plant at the end of the drought.

Selection	Internode length index	Stolon elongation rate index	Percent of branching nodes	Number of branches	Percentage of nodes rooted index	Total clover dry weight index
<u>Cultivar</u>						
Kopu	0.96	0.65	0.01	0.05	0.51	0.70
Tahora	0.70	0.46	0.01	0.04	0.74	0.40
<u>Internode length</u>						
Long	0.75	0.48	1.3	0.06	0.60	0.81
Short	1.0	0.55	2.6	0.08	0.57	0.92
<u>Percentage of nodes branching</u>						
High	0.72	0.47	0.0	0.0	0.52	0.24
Low	0.82	0.71	3.0	0.03	0.80	0.33
<u>Light</u>						
Sun	0.96	0.96	2.0	0.07	0.68	0.67
Shade	0.71	0.71	0.4	0.03	0.56	0.43
<u>Contrast</u>						
Kopu vs. Tahora	NS	NS			NS	NS
Long vs. Short internode length	NS	NS			NS	NS
High vs Low % of nodes branching	NS	NS			NS	NS
Sun vs. Shade	*	*			NS	*

NS = not significant, * p < or = 0.05, ** p < or = 0.01

Table 2

The mean recovery index for plant characters across all cultivars selections for each selectio, with the significance of pre-planned contrasts.

Selection	Internode length index	Stolon elongation rate index	Percent of branching nodes	Number of branches index	Percentage of nodes rooted index	Clover dry weight index
<u>Cultivar</u>						
Kopu	1.1	1.2	0.25	1.3	1.0	1.9
Tahora	1.0	1.0	0.36	2.1	0.90	1.3
<u>Internode length</u>						
Long	1.1	1.0	0.27	1.4	1.0	1.6
Short	1.1	1.0	0.34	2.0	0.99	2.6
<u>Percentage of nodes branching</u>						
High	0.87	0.94	0.37	2.2	1.0	0.93
Low	1.0	1.0	0.23	1.3	1.0	1.3
<u>Light</u>						
Sun	1.1	1.0	0.35	2.0	1.0	1.0
Shade	0.96	0.99	0.27	1.5	0.97	1.4
<u>Contrast</u>						
Kopu vs. Tahora	NS	NS	NS	NS	NS	NS
Long vs. Short internode length	NS	NS	**	**	NS	NS
High vs Low % of nodes branching	NS	NS	NS	NS	NS	NS
Sun vs. Shade	NS	NS	NS	NS	NS	NS

NS = not significant, * p < or = 0.05, ** p < or = 0.01