

## Effect of summer moisture deficit on growth of five white clover cultivars

X. WANG\*, J.R. CARADUS<sup>2\*</sup> and A.C.P. CHU<sup>1</sup>

<sup>1</sup> Plant Science Department, Massey University Private Bag, Palmerston North

<sup>2</sup> AgResearch Grasslands, Private Bag 11008, Palmerston North

### Abstract

Growth of five New Zealand white clover cultivars, Grasslands Kopu, Grasslands Pitau, Grasslands Huia, Grasslands Tahora and Prop, was quantified at differing soil moistures in both the field and the glasshouse. The first trial employed a rain-out shelter to impose two soil water treatments. While there were no differences among the cultivars for leaf water status, there were differences in plant growth parameters in response to water deficit. Water deficit did not significantly affect leaf appearance rate of Prop, although there was a 2-fold difference. There was, however, a 3.5- to 6-fold decrease in leaf appearance rate due to water deficit for the other cultivars. Water deficit did not significantly reduce leaf size for the medium- and small-leaved cultivars Huia, Tahora and Prop; but was halved for Kopu and Pitau. Prop had the lowest stolon growing point survival under water deficit and Pitau the highest. Leaf longevity was greatest for Prop and least for Pitau when grown under optimum water supply, but this pattern was reversed under water deficit stress. The second trial, a pot trial, investigated the response of the same five cultivars to three different soil water regimes (control, mild and severe stress). The growth parameters of smaller-leaved cultivars, particularly Prop, were less affected than the large-leaved cultivars in their response to water deficit. These short-term trials showed that some small-leaved cultivars of white clover have an ability to adjust their growth and habit in response to water deficit more effectively than large-leaved cultivars. Prop was able to maintain a higher leaf appearance rate than other cultivars when grown under water deficit. However, while exhibiting this drought tolerance adaptation the low stolon growing point survival of Prop could result in a poor recovery from drought. Small-leaved cultivars are rarely tap-rooted, a characteristic of plants adapted to more prolonged drought conditions. The probability of combining these characteristics and improving summer production of white clover through identification of drought tolerance is discussed.

**Keywords:** cultivars, drought, *Trifolium repens*, variation, water deficit

### Introduction

Most of New Zealand's pastoral area has an average annual rainfall between 750 and 2000 mm. While this is adequate for temperate pasture production, water deficits may occur in many areas during summer, owing to high evapotranspiration. White clover (*Trifolium repens* L.) can complement spring perennial ryegrass growth by contributing relatively more to summer herbage production than in other seasons (Brougham 1966). However, this growth potential will be reduced by water deficit stress which not only slows growth but in extreme cases also reduces the persistence of white clover (Thomas 1984). White clover cultivars that can withstand water deficit stress are a priority, and management strategies that can reduce the detrimental effect of water deficit stress on white clover growth will together help to enhance summer production.

Some morphological characteristics have been related to plant stress tolerance. Tap-rooted white clovers have, in some environments, been considered drought tolerant (Smith & Morrison 1983). Small-leaved types, which have high stolon growing point density, but are rarely tap-rooted, are known to persist in less favourable environments (Caradus & Williams 1989; Brock & Kim 1994). Profuse and/or early flowering may also be an advantage in certain adverse environments, such as severe summer drought (Jones 1980; Macfarlane & Sheath 1984).

The objective of this study was to identify and, if possible, quantify morphological and growth differences between five New Zealand white clover cultivars grown under different levels of soil water in both the field and controlled environment facilities.

### Materials and methods

#### Plant material

The five white clovers evaluated were Grasslands Kopu, Grasslands Pitau, Grasslands Huia, Grasslands Tahora and Prop.

\* Corresponding author.

### Field trial

The field trial was located at the Pasture and Crop Research Unit, Massey University, Palmerston North on a Tokomaru silt loam soil (Cowie *et al.* 1972) and a 3-year-old perennial ryegrass-white clover pasture. In August 1985 the area was sprayed with paraquat and ploughed 10 days later. Lime (3 t/ha) and NPK (5: 10: 12) compound fertiliser were incorporated into the soil by rotary cultivation. In September, seedlings were transplanted into the site at 50 cm centres and tagged with a 120 mm plastic ring. The area between white clover plants was sown with Grasslands Nui perennial ryegrass. The plots were mown every 8 weeks until the start of water stress treatments.

Before water treatments were imposed on 1 December 1985 all plots were irrigated to field capacity. The two water treatments, imposed for 75 days during summer, were (a) water stress, where plots were covered by 2 automatic rain-out shelters (each 10 × 5 m) to create a gradually increasing water deficit from 25 mm to 171 mm, (b) unstressed, where water deficit remained between 23 and 31 mm by irrigating according to the model of Scotter *et al.* (1979). There were 2 plots under the rainout shelter and 2 with irrigation for each cultivar. For each plant 3 primary stolons with at least 8 nodes were tagged with coloured wires on the internode after the youngest emerged node. Measurements were:

- (a) soil volumetric water content, by gravimetric sampling to 50 cm depth fortnightly and adjusting for a soil bulk density of 1.15;
- (b) relative water content (RWC) of the youngest fully expanded leaf between 1200 and 1300 hours every 15 days;
- (c) leaf appearance rate (LAR) measured as number of leaves produced from the tagged position;
- (d) leaflet length of 12 fully expanded leaves per plant grown during the stress period, at the end of the trial;
- (e) stolon growing point survival by counting the growing points on the 3 tagged stolons of each plant every 2 weeks during the last month of the trial; and
- (f) leaf longevity derived from dividing the number of leaves per stolon by leaf appearance rate.

Data were analysed using the pooled environment model. Within each environment the cultivars were arranged in a randomised block design, allowing separate analysis of cultivar effect, treatment effect and their interactions. The data were analysed using ANOVA in the statistical package for social sciences (SPSS).

### Glasshouse trial

The experiment was carried out in the Plant Growth Unit, Massey University, Palmerston North from March to May 1986, during which time temperatures were maintained between 18 and 25°C. Pots (4 l) contained a peat:pumice:sand medium (70:20:10 by volume basis) into which a compound fertiliser (P, K, S, lime and micronutrients) was mixed.

White clover plants were vegetatively propagated from those used in the field in February 1985. After growing for 8 weeks the plants were defoliated and cut to a uniform size of about 5 main stolons.

A completely randomised block design was used with treatments being 5 white clover cultivars and 3 water levels (control = 30% (g/100 g) soil water content (SWC); mild stress = 17-20% soil water content; severe stress = 4% soil water content) and 5 replicates. The moisture treatments were maintained by adding water equivalent to the amount lost daily, calculated by weighing 5 sample pots per treatment.

Measurements were:

- (a) leaf appearance rate estimated from the number of leaves produced subsequent to a marked internode at the third youngest node of a stolon which had at least 8 nodes, after 60 days;
- (b) petiole extension rate estimated from measurements of petiole length measured on the most recently emerged leaf every day for 5 days;
- (c) leaf RWC measured by the same method as described for the field trial, on 4 replicates in 4 periods (days 0 to 7, days 20-27, days 40-47 and days 60-67); and
- (d) total plant (shoot and root) dry weight, after 80 days.

### Results

In the field trial, leaf RWC was between 90 and 92% for the unstressed control treatment; but decreased from 92% to 55% in the water deficit treatment during the 75-day period. There were no significant differences among cultivars for leaf RWC.

In the pot trial, there was also no significant difference among cultivars for leaf RWC at any of the water deficit levels (means were 94%, 71% and 51% with increased water deficit). Total plant dry weight after 80 days was reduced to 23% and 4% of control plants at moderate and severe water stress levels, respectively. There was no significant difference between cultivars for total plant dry weight at any water level or a significant cultivar × water level interaction.

In both trials LAR was less affected by water deficit for Prop than the other cultivars (Tables 1 and 2). In the

glasshouse under severe water deficit LAR of Pitau was significantly less than for all other cultivars (Table 2). Water deficit significantly reduced leaf size of Kopu, Pitau and Huia unlike the smaller-leaved Tahora and Prop (Table 1). Petiole extension rate was less affected by water deficit for Prop than the other cultivars in the glasshouse trial (Table 2).

Stolon growing point survival was similar for all cultivars when unstressed but significantly lower for Prop compared with Kopu, Pitau and Huia when grown under water deficit (Table 1). Leaf longevity of Prop was significantly greater than for other cultivars when grown with adequate water supply, but along with Huia and Tahora had significantly lower leaf longevity than Kopu and Pitau when grown under water deficit (Table 1).

## Discussion

Although there were differences among cultivars for many of the plant characters measured, these may not necessarily ensure drought tolerance. Both dry matter accumulation and RWC showed little variation among cultivars grown either with or without water stress. It could be argued that since these are critical indicators of drought tolerance few differences in drought tolerance between cultivars exist. Despite this, some of the inter-cultivar differences identified, in these short-term trials,

may have more profound effects in perennial swards. The most noticeable differences among cultivars were that Prop, and to a lesser extent Tahora, had a more plastic response for stolon growing point survival to water deficit than the other cultivars (particularly the larger-leaved cultivars), but a less plastic response for leaf appearance rate, leaflet length and petiole extension rate. Prop was selected from populations collected from summer dry hill country (Macfarlane & Sheath 1986). In a study using larger soil-filled pots with a continuous dry-down period over 37 days (i.e., there were no wetting/drying cycles) a white clover line from Syria, which had exhibited some drought tolerance (Caradus et al. 1990a) had a higher leaf appearance rate than Kopu, suggesting that this Syrian ecotype had the ability to continue growth into dry conditions (Barker et al. 1993). In the current study Prop also had a significantly higher leaf appearance rate than Kopu but only when grown under water deficit (Tables 1 and 2). With adequate water supply leaf appearance rate of Prop significantly was lower than all other cultivars in the field and for Kopu in the glasshouse trial.

White clover stolons adjust osmotically and survive longer than leaves when subjected to water deficit (Turner 1991) so that number of plants surviving drought can be related to pre-drought plant density or stolon growing point number (Brock & Kim 1994). Prop had lower stolon growing point survival when subjected to

Table 1 Leaf appearance rate, leaflet length, stolon growing point survival, and leaf longevity of white clover cultivars grown under 2 moisture levels in the field.

Cultivar	Leaf appearance rate (leaves/wk)		Leaflet length (mm)		Stolon growing point survival (%)		Leaf longevity (days)	
	Unstressed	Stressed	Unstressed	Stressed	Unstressed	Stressed	Unstressed	Stressed
Kopu	1.17	0.19	20	11	95	79	34	53
Pitau	1.17	0.19	15	7	98	91	26	63
Huia	0.99	0.28	13	a	92	75	34	43
Tahora	1.17	0.22	10	a	97	73	30	38
Prop	0.77	0.30	9	a	99	64	43	37
P (main effect)	*	ns	**	*	NS	*	*	*
LSD <sub>0.05</sub>	0.13		3	3			7	10
P (interaction)		*		*		*		*
LSD <sub>0.05</sub>		0.52		4		15		25

Table 2 Leaf appearance rate and leaf petiole extension rate of white clover cultivars grown at 3 moisture levels in pots, in a glasshouse.

Cultivar	Leaf appearance rate (leaves/wk)			Petiole extension rate (mm/day)			
	Unstressed	Moderate stress	Severe stress	Unstressed	Moderate stress	Severe stress	Severe stress
Kopu	1.29	0.70	0.49	11.4	3.5	0.1	
Pitau	1.59	0.56	0.21	12.2	5.6	0.1	
Huia	1.52	0.56	0.35	5.4	1.2	0.2	
Tahora	1.59	0.50	0.35	4.9	2.2	0.0	
Prop	1.25	1.19	0.70	3.4	2.1	0.5	
P	*	*	**			*	
LSD <sub>0.05</sub>	0.61	0.16	0.10	2.8	3.2	0.5	

water deficit than other cultivars (Table 1), but was able to maintain leaf appearance rate to a greater extent than other cultivars during water deficit (Table 2). Therefore, while Prop may tolerate drought by maintaining leaf appearance rate its low stolon growing point survival could result in poorer recovery from drought.

Larger tap-root diameter has been associated with populations of white clover from dry sites (Caradus & Woodfield 1986; Caradus 1991), and may be considered an adaptive character. The close genetic association between leaf size and tap-root diameter (Woodfield & Caradus 1990) could mean that combining a medium-small leaf type similar to that found for white clovers from grazed dryland sites (Caradus *et al.* 1990b) with a larger tap-root may prove elusive. Dual selection for larger tap-root diameter and medium-small leaf size has been moderately successful (Woodfield & Caradus 1987, 1990). Selection for tap-rootedness per se has proved of some benefit for drought tolerance in field trials when selections were made within dryland populations (van den Bosch *et al.* 1993).

Adaptive characters that may allow improved tolerance to drought could include a higher (or maintained) leaf appearance rate and tap-rootedness.

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