RESPONSE OF THE ONTOGENY, YIELD, AND PHOSPHORUS NUTRITION OF GRASSLANDS TAHORA AND GRASSLANDS HUIA WHITE CLOVER TO PHOSPHORUS

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ABSTRACT

The ontogeny, morphology, yield, and P nutrition of Tahora and Huia white clover were compared under P deficient and P sufficient conditions in a glasshouse pot trial. The total and relative yield of Tahora was greater than that of Huia under P deficient conditions. The number of stolons per plant was greater for Tahora than Huia in both absolute and relative terms under P deficiency. The proportional increase in root yield was similar for both cultivars as was the decrease in the rate of ontogeny under P deficiency. The P nutrition of both cultivars in terms of net P uptake and P concentrations in tissues was similar. The major difference between Tahora and Huia was the greater sensitivity of Huia stolons to P deficiency.

INTRODUCTION

The search for a P efficient white clover selection has been long and difficult (Dunlop *et al.*, 1988). At the micro level of P efficiency research it has been difficult to both define and discern attributes that confer P efficiency on white clover (Dunlop *et al.*, 1988; Hart, 1988). The efficiency of P uptake by white clover has been related to root size, measured as weight or length, (Hart *et al.*, 1981; Caradus, 1983) but the efficiency of P utilisation by white clover has been related to a wider group of factors. For example, P utilisation has been shown to be affected by the proportional allocation of P between the inorganic-P, ester-P, lipid-P, and residual-P fractions, the area of compartmentation of the P fractions within the plant cell, and the demands of nitrogen fixation in nodulated plants (Chisholm & Blair, 1988b; Hart, 1988).

On the other hand research at the macro or ecological level has demonstrated differences in the productivity and persistence of white clover selections grown on low P soils that were related to root and stolon morphology rather than the commonly used micro definitions of P efficiency (Williams & Caradus, 1979; Simpson *et al.*, (1987).

Clearly, the response of white clover plants to P deficient conditions includes changes in P acquisition, P utilisation, and plant morphology. In addition stress, such as P stress, can disrupt the course of a plant's ontogeny or the rate of its ontogeny (Grime *et al.*, 1986). Therefore, the objective of this experiment was to compare the response of the ontogeny, morphology, biomass yield, and P nutrition of two cultivars of white clover under P deficient conditions.

MATERIALS AND METHODS

The two cultivars of white clover (*Trifolium repens* L.) used were Grasslands Tahora and Grasslands Huia, hereafter referred to as Tahora and Huia. The seeds were sown in soil and immediately after emergence of the cotyledons a single plant was transplanted into each pot. The top of each pot was covered by heavy duty black plastic except for a 20 mm hole in the centre of the plastic into which the plant was transplanted. The plastic cover on the pot prevented the growth of roots from stolon nodes and was used to prevent variations in root production and stolon morphology that usually result from a varying number of rooted nodes developing on white clover plants growing in pots. The pots were placed in dishes and watered daily to field capacity from the bottom of the pot.

The two P rates used were 200 ppm and 2,000 ppm P w/w applied as H_3PO_4 and mixed through the soil. Hereafter these rates are referred to as the low P rate (LP) and the high P rate (HP). These P rates were shown by Hart & Jessop (1983) to create P deficient conditions and P sufficient conditions in the Egmont sandy loam soil (1.9 kg/pot) used (New Zealand Soil Bureau, 1968).

All plants were inoculated with rhizobia. The nutrients, and the rates used (g/kg dry soil) mixed into the soil were as follows: $MgSO_4.7H_2O$ (0.6), K_2SO_4 (0.9) and $Na_2MoO_4.2H_2O$ (0.001)

The experiment was conducted in a glasshouse from October to December with the day temperature set at 23 °C and the night temperature at 15 °C. The extreme temperature range during the experiment was from 10 to 28 °C.

The experimental design was a randomised complete block with four replicates, two cultivars, two P rates and four harvests. The harvests were at 26, 41, 47, and 69 days after transplanting (DAT). The harvest times were chosen to cover the developmental stages of white clover between early vegetative growth and early flowering.

At each harvest the number of leaves, axillary buds, lateral branches, lateral stolons, main stolons, and flowers were counted on the basis of the definitions of Thomas (1987). These data were summarized in an ontogenetic scale specific to this experiment (Table 1) that was based on the decimal coded system of Zadoks *et al.* (1974).

Table 1: A decimal coded ontogenetic scale for white clover.

Number of leaves	
10	Number of axillary buds
20	Number of lateral branches
30	Number of lateral stolons
40	Number of main stolons
50	Flower formation visible
51	Green flowerheads
55	White flowers in 50 % of flowerheads
60	Seed formation

The dry weight yield and the P concentration of the roots, stolons and leaves plus petioles were also measured at each harvest. The length of the lateral and main stolons on each plant was also recorded. The P concentration was analysed by the method of Haslemore & Roughan (1976).

The data were analysed by analysis of variance and only significant interactions and main effects are presented. The heterogeneity of variance common in time course experiments with plants resulted in the use of a $\log_{10}$ plus one transformation. All means are presented as detransformed means.

RESULTS

Ontogeny of shoots: The rate of development of the shoots of both Huia and Tahora was slower at 200 ppm P than at 2,000 ppm P, but there was no significant difference in the ontogenetic stage of Huia and Tahora at either P rate at any of the harvest times (Table 2). Tahora was observed to produce flowers earlier at the high P rate (2,000 ppm P) than at the low P rate (200 ppm P) and also to produce flowers earlier than Huia (see Table 2). There were more main stolons per plant on Tahora than Huia at the high P rate (7 vs 5) at 69 DAT and at the low P rate some Tahora plants produced main stolons but there were no main stolons on the Huia plants (unpublished data).

Table 2: The decimal coded ontogeny of white clover over time at high and low P rates.

	LP		HP	
DAT	Huia	Tahora	Huia	Tahora
26	2.0g*	4.0g	12.8f	12.8f
41	14.0ef	17.3e	33.5bc	33.5bc
47	21.8d	27.5cd	34.8b	39.3b
69	33.5bc	38.0b	45.0ab	55.0a

* Means without a letter in common are significantly different ($P < 0.05$).

At the low P rate there were eleven more branches per plant on Tahora than on Huia at 69 DAT but there was no significant difference between the two cultivars in the number of branches per plant at the earlier harvest times (Table 3). The major contributors to the greater number of branches per plant on Tahora at the low P rate were axillary and lateral buds. At the high P rate the number of branches per plant for Huia and Tahora was similar at 69 DAT but was significantly greater than the number of branches per plant for either cultivar at the low P rate (Table 3).

Biomass allocation: The total dry weight per plant of Tahora was significantly greater than that of Huia at the low P rate over all harvest times (Table 4). The cultivar by P rate by harvest time interaction was not significant. At 69 DAT the total dry weights per plant of Tahora and Huia at the low P rate were 1.757 g and 0.890 g respectively. The total dry weight per plant at the low P rate for Tahora was 18 % of the yield at the high P rate whereas the corresponding value for Huia was 9 % (Table 4). There was no significant difference in the total dry weight per plant of Tahora and Huia at the high P rate (Table 4).

Table 3: The number of branches on white clover over time at high and low P rates.

	LP		HP	
DAT	Huia	Tahora	Huia	Tahora
26	0.00g*	0.00g	2.75ef	0.00g
41	2.00f	2.75ef	12.00d	16.75c
47	3.25e	4.00e	22.00bc	30.00b
69	15.5cd	26.5b	99.5a	98.5a

* See Table 2.

The cultivar by time and the cultivar by P rate by time interactions were not significant for the dry weights of leaf plus petiole, stolon, and roots. For this reason and the minimal stolon production at the earlier harvest times the component dry weights were analysed separately for 69 DAT (see Table 5).

Table 4: The total dry weight per plant (g/plant) of white clover at high and low P rates.

Cv	LP	HP
Huia	0.275c*	2.972a
Tahora	0.507b	2.886a
DAT	**LP**	**HP**
26	0.015f	0.063ef
41	0.053ef	0.875d
47	0.173e	1.614b
69	1.324c	9.166a

* See Table 2.

At the low P rate the dry weight yields of leaf plus petiole, stolon, and roots of Tahora were significantly greater than those of Huia whereas at the high P rate only the stolon dry weight of the two cultivars was significantly different (Table 5). The relative proportion that the yield components leaf plus petiole, stolon, and roots contributed to total plant yield was different at the two P rates. At the high P rate the total plant dry weight was 36 and 26 % stolon for Tahora and Huia respectively where as the respective proportions for Tahora and Huia at the low P rate were 17 and 11 % (Table 5). Conversely, the proportion of total plant dry weight as root increased from 19 % at the high P rate to 32 % at the low P rate for Tahora and from 20 % to 35 % for Huia (Table 5). The proportion of total plant yield that was leaf plus petiole was relatively insensitive to P rate and was approximately 50 % at both P rates for both cultivars (Table 5).

Table 5: The leaf plus petiole, stolon, and root dry weight of white clover (g/plant) dry weight of white clover (g/plant) at low and high P rates 69 days after transplanting.

Cv	Leaves	Stolon	Roots
		LP	
Huia	0.479c*	0.096d	0.315c
Tahora	0.891b	0.298c	0.567b
		HP	
Huia	4.930a	2.332b	1.860a
Tahora	4.166a	3.342a	1.702a

* Means within each column without a letter in common are significantly different ($P < 0.05$).

The total length of main stolons and lateral stolons at the low P rate at 69 DAT on Tahora was 40.5 cm which was significantly greater than the 16.3 cm of stolon on Huia. The total stolon lengths of Tahora (142.1 cm) and Huia (140.9 cm) were similar at the high P rate at 69 DAT. Similarly to stolon dry weight the total stolon length of Tahora was greater at the low P rate relative to its stolon length at the high P rate than was Huia.

Nutrition: There was no significant difference in the net P uptake (i.e. total P content) of Huia and Tahora when compared at any P rate or harvest time. Over all harvest times and P rates the net P uptake of Huia was 0.85 mg P/plant and that of Tahora 0.77 mg P/plant. The net P uptake of both cultivars was greater at the high P rate than the low P rate at each harvest time (Table 6).

Table 6: The net P uptake (mg P) of white clover plants at high and low P rates

DAT	LP	HP
41	0.097e*	5.180c
47	0.356e	8.493b
69	3.294d	36.344a

* See Table 2.

Tahora allocated a greater proportion of the P content of the plant to the stolons than Huia at both the low and the high P rate (Table 7). The allocation of P to the stolons of both cultivars was in proportion to the allocation of biomass to the stolons (cf Tables 5 and 7).

Table 7: The leaf plus petiole, stolon and root P content of white clover (mg P/plant) at low and high P rates 69 days after transplanting.

Cv	Leaves	Stolon	Roots
		LP	
Huia	1.6b*	0.2c	1.2b
Tahora	1.7b	0.6c	1.8b
		HP	
Huia	17.5a	9.5b	10.1a
Tahora	14.0a	13.2a	8.4a

* See Table 5.

The P concentration of the leaves, stolons, and roots of Tahora and Huia were not significantly different at any P rate or harvest time. The mean concentrations for both cultivars over all P rates and harvest times were 0.32, 0.28, and 0.47 % for the leaves, stolons, and roots respectively. The concentration of P in the leaves, stolons, and roots was greater at the high P rate than the low P rate for all harvest times when meaned over both cultivars (unpublished data).

DISCUSSION

The total plant yield of Tahora was greater than that of Huia under the P deficient conditions of the low P rate (Table 4). At 69 DAT the yield components leaf plus petiole, stolon, and root dry weight were all greater in Tahora than Huia at the low P rate. In contrast, Hart (1986) found no difference in the plant dry weights of Tahora and Huia under P deficient conditions. The plants in Hart's (1986) experiment were regularly defoliated and the stolons were allowed to root at the nodes. Presumably the confounding effect of defoliation on dry weight yield response to P level resulted in the conflict between the results reported here and those of Hart (1986).

The plastic response of Huia and Tahora to P deficiency was similar although the degree of the response, for the stolons in particular, was different. In both cultivars of white clover the allocation of total biomass to the three vegetative yield components at the low P rate relative to the high P rate resulted in proportionally lower stolon yields proportionally higher root yields and a proportionally similar leaf plus petiole yield (see Table 5). Whereas a proportional increase in the total biomass allocated to roots, or an increase in the root:shoot ratio, under P deficient conditions has often been reported (e.g. Hart *et al.*, 1981; Kemp, 1985). The sensitivity of the proportion of total plant biomass allocated to stolons under P deficient conditions has been less widely recognised (Hart, 1986; Dunlop *et al.*, 1988).

In terms of defining P efficient white clover plants the relatively greater sensitivity of the stolon yield of Huia relative to Tahora would appear to be worthy of further study. Tahora was selected for its stoloniferous growth habit (Caradus & Williams, 1981) yet in this trial it was only at the low P rate that Tahora exhibited greater number of stolons in absolute, and relative, terms (Table 3). This greater insensitivity of the stolons on Tahora to P deficiency, compared with Huia, would confer a greater ability to forage bare ground, and thereby the opportunity to grow roots into soil containing P, on Tahora. Grime (1979) has defined the plastic morphology of white clover as being a competitive strategy but clearly within the species of white clover the degree of plastic morphology with respect to stolons, and therefore the competitiveness of the plants, varies between selections.

The greater yield of Tahora relative to that of Huia under P deficient conditions was driven by the greater number of stolons and stolon buds on Tahora and the resultant greater stolon yield (Tables 2,3,5). A consequence of the greater stolon yield of Tahora was the greater proportion of the plant P content in the stolons. As it is the leaf plus petioles rather than the stolons that are removed during grazing (Jewiss, 1981) more P would be retained for remobilisation into regrowth in plants that are more stoloniferous (see Caradus & Williams, 1981).

There was no evidence that there were P efficiency differences between Huia and Tahora other than the morphological response of the stolons. The net P uptake and the P concentrations of roots, stolons, and leaves plus petioles were similar for both cultivars (Tables 6,7). Hart (1986) also found no difference between the P nutrition of Tahora and Huia.

Overall, P deficiency affected white clover by slowing the rate of ontogeny, as evidenced by the delayed onset of stolon production and flowering, by affecting the number of growth units, as evidenced by the decreased number of stolon branches, but not by disrupting the course of the plant's ontogeny. Nevertheless, the degree of the plastic response to P deficiency of Huia and Tahora differed particularly the response of stolon morphology and stolon yield. Finally, these results demonstrate the value of interpreting the response of white clover to P deficient conditions, or other stresses, in terms of changes in the ontogeny, morphology, biomass production, and P nutrition of the plant rather than by considering components of the plants growth and development in isolation.

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REFERENCES

Caradus, J.R. & Williams, W.M. 1981. Breeding for improved white clover production in New Zealand hill country. In *Plant physiology and herbage production*. (ed. Wright, C.E.). pp 163-168, *Occasional symposium No. 13, British Grasslands Society.*

Chisholm, R.H. & Blair, G.J. 1988b. Phosphorus efficiency in pasture species. II. Differences in the utilisation of P between major chemical fractions. *Australian Journal of Agricultural Science* ***39***, 817-826.

Dunlop, J., Wewaka, G.S., Caradus, J.R., Mackay, A.D., Lambert, M.G., Hart, A.L., Bosch, J. van den, Mouat, M.C.H. & Hay, M.J.M. 1988. A Grasslands Division programme to breed a white clover with improved phosphate nutrition. *Proceedings of the Agronomy Society of New Zealand* ***18***, 71-74.

Grime, H.P. 1979. *Plant Strategies and Vegetation Processes.* John Wiley, Chichester.

Hart, A.L. 1986. A comparison of the response to phosphorus of *Trifolium repens* 'Grasslands Huia' and 'Grasslands Tahora' in a controlled environment. *New Zealand Journal of Agricultural Research* ***29***, 179-182.

Hart, A.L. 1988. The physiology of phosphorus and its relevance to phosphorus efficiency in white clover. *Proceedings of the Agronomy Society of New Zealand* ***18***, 37-40.

Hart, A.L., Halligan, G. & Haslemore, R.M. 1981. Analysis of the response of pasture legumes to phosphorus in a controlled environment. *New Zealand Journal of Agricultural Research* ***24***, 197-201.

Hart, A.L. & Jessop, D. 1983. Phosphorus fractions in trifoliate leaves of white clover and lotus at various levels of phosphorus supply. *New Zealand Journal of Agricultural Research* ***26***, 357-361.

Haslemore, R.M. & Roughan, P.G. 1976. Rapid chemical analysis of some plant constituents. *Journal of the Science of Food and Agriculture* ***27***, 1171-1178.

Jewiss, O.R. 1981. Shoot development and number. In *Sward Measurement Handbook*. (ed. Hodgson, J. *et al.*). pp 93-113. British Grassland Society, Hurley.

Kemp, P.D. 1985. The efficiency of acquisition and utilisation of phosphate by four temperate pasture species. PhD thesis, University of New England, Armidale.

New Zealand Soil Bureau 1968. Soil Bureau Bulletin 26.

Simpson, D., Wilman, D. & Adams, W.A. 1987. The distribution of white clover (*Trifolium repens* L.) and grasses within six sown hill swards. *Journal of Applied Ecology* ***24***, 201-216.

Thomas, R.G. 1987. The structure of the mature plant In *White Clover*. (ed. Baker, M.J. & Williams W.M.). pp 1-30. CAB International, Wallingford.

Williams, W.M. & Caradus, J.R. 1979. Performance of white clover lines on New Zealand hill country. *Proceedings of the New Zealand Grassland Association* ***40***, 162-169.

Zadoks, J.C., Chang, T.T. & Konzak, C.F. 1974. A decimal code for the growth stages of cereals. *Weed Research* ***14***, 415-421.