Defoliation Effects on Root and Rhizome Development of Kura Clover

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ABSTRACT : There is limited information on the relationship between defoliation and root and rhizome development of kura clover (*Trifolium ambiguum* M. Bieb.). To determine the effects of defoliation severity on root and rhizome growth of young kura clover plant (seedling about 8 wk old), this research was conducted in 2002 (Experiment 1) and 2003 (Experiment 2) in a glasshouse at the University of Wisconsin-Madison. Four kura clover entries were used in this experiment: two were started from seed materials (ARS-2678 and 'Rhizo') and two were clones from mature, field-grown Rhizo kura clover plant. Three defoliation frequencies (2-, 4- and 6-wk intervals) and two defoliation intensities (complete and partial defoliation) were imposed on each of the four kura clover entries. Root, rhizome, and leaf dry matter (DM) generally increased with less frequent defoliation, however, the increase in rhizome DM was not significant between 4- or 6-wk defoliation periods. The root and leaf DM under complete defoliation (CD) were significantly lower than under partial defoliation (PD). In Exp. 1, rhizome DM was not significantly different between CD and PD; it was significantly lower under CD in Exp. 2. ARS-2678 showed excellent root development characteristics, however, its rhizome DM was significantly lower than Rhizo clones. The rhizome development from Rhizo clones was greater than that from seed materials. If maximum root and rhizome growth are expected from young kura clover plant, the intensity and frequency of defoliation should be minimized or defoliation should be avoided. ***(Asian-Aust. J. Anim. Sci. 2006. Vol 19, No. 5 : 690-694)***

Key Words : Kura Clover, Rhizome, Root

INTRODUCTION

Defoliation has been shown to have a substantial impact on root, stolon, and rhizome development in various forage species. A number of papers (Evans, 1973; Halliday and Pate, 1976; Ryle et al., 1985; Sheaffer et al., 1988) have described frequent defoliation as having a negative impact on below-ground growth, including depletion of root carbohydrate reserves. The length of the stolon internodes of white clover increases with long periods without defoliation (Grant and Barthram, 1991). Ryle et al. (1985) indicated that the rate of N_2 fixation was reduced by more than 70% under defoliation.

Kura clover (*Trifolium ambiguum* M. Bieb) has a rhizome system, which enables it to spread extensively under some conditions. Kura clover persistence is attributed to these unique organs not found in other commercially useful clovers adapted to the temperate region. Kura clover is considered to have potential as a pasture legume because of its great persistence under environmental extremes. It has good winter hardiness, is adapted to diverse soil types, tolerates frequent defoliation, and tolerates drought (Spencer et al., 1975; Speer and Allinson, 1985; Peterson et al., 1994). Kura clover is more tolerant of poor drainage than red clover (*Trifolium pratense* L.) or white clover (*Trifolium repens* L.) and persists through drought by becoming dormant (Speer and Allinson, 1985; Bryant, 1997). Unique botanical characteristics of kura clover, such as vegetative propagation by means of rhizomes, should improve the grazing persistence of kura clover compared to other legumes (Taylor and Meche, 1982). Speer and Allison (1985) reported that kura clover has multi-branched roots that consistently extend to a depth exceeding 60 cm, and kura clover started from rhizome segments spreads laterally in excess of 1.2 cm by early in the second season of growth. They reported that rhizome development initiated approximately 3 months after seeding, and daughter plants developed within 45 days after planting single-node rhizome segments, even in soil with a pH as low as 4.8. Peterson et al. (1994) showed that clipping treatments ranging from three to six cuttings per year had little effect on a 5-year-old kura clover below-ground growth.

Knowledge about the relationship between defoliation and rhizome development of kura clover, particularly several defoliation regimes, is lacking but essential to need. It is important to understand the growth dynamics of the rhizome system of kura clover since establishment to make decisions on defoliation of kura clover in pasture situations. The objective of this study was to determine the effects of defoliation severity on root and rhizome development of young kura clover plants.

MATERIALS AND METHODS

This research was conducted in 2002 (Exp. 1) and 2003 (Exp. 2) in a glass house on the University of Wisconsin-Madison campus. Four kura clover entries were used in this experiment: two were started from seed (ARS-2678 and

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Table 1. The sources and characteristics of four kura clover entries used in 2002 and 2003

Population	Entry	Source	Characteristics
	ARS-2678	Utah Agricultural Exp. Station	Spreading extensively
Rhizo		USDA-SCS and Univ. of Kentucky	Spreading extensively
Clone	R1	Rhizo	Spreading with long rhizomes
	R2	Rhizo	Spreading with medium rhizomes

Table 2. Description of defoliation intensity, defoliation frequency, and combined defoliation intensity and frequency treatment (DIF) imposed on each kura clover entry

Treatment	Description
Intensity	
CD	Complete defoliation: removal of all plant herbage
PD	Partial defoliation: removal of all plant herbage except for the two most recently expended leaves
Frequency	
2-wk	Cut every 2 week
4-wk	Cut every 4 week
6-wk	Cut every 6 week
DIF[1]	
2-wk CD	Cut every 2 week with complete defoliation
4-wk CD	Cut every 4 week with complete defoliation
6-wk CD	Cut every 6 week with complete defoliation
2-wk PD	Cut every 2 week with partial defoliation
4-wk PD	Cut every 4 week with partial defoliation
6-wk PD	Cut every 6 week with partial defoliation

[1] A created treatment whose value is the combination of values of defoliation frequency and defoliation intensity.

‘Rhizo’) and two were clones from mature, field grown Rhizo kura clover plants. Clones were developed from uniform two-mode segments of two different Rhizo plants. The sources and characteristics of plant materials used are described in Table 1.

Ninety six pots were filled with a mixture of one part each sand, peat moss, and vermiculite to two parts soil, in preparation for seeds and for rhizome transplants. Two rhizomes or two seeds of each of the four kura clover entries were planted in each pot and were thinned to one plant per pot after 2 weeks. Nutrient solution including P, K, Mg, K, Ca and micronutrients were applied to each pot one month after planting and shortly after the first defoliation treatment.

Metal halide lamps (400 W) were used to supplement natural light. The lamps were turned on for 16 hr per day (5 am through 9 pm) with the temperature of the greenhouse kept at 21°C at night and at 25°C during the day to simulate the growing season, the experiments were conducted from January through June in 2002 and in 2003.

Defoliation intensity and defoliation frequency treatments were imposed on each of the four kura clover entries. A factorial treatment design was used as follows: three defoliation frequencies (2-, 4- and 6-week intervals)× two defoliation intensities (complete and partial defoliation) ×four different kura clover entries. Partial defoliation involved removal of all but the two most recently expended leaves. Pots were arranged in a completely randomized block design with four replications in each of 2 years. Treatments are described in Table 2.

Defoliation treatments were initiated 2 months after planting and each treatment lasted 12 weeks. The initial defoliation took place on the same day for all treatments and the 2-, 4-, and 6-week defoliations were conducted 6, 3, and 2 times, respectively, during the 12-week period. Herbage yield at each harvest was measured after it was dried at 60°C. This measurement was taken to obtain information about the above ground growth response to defoliation. At the conclusion of the experiment (12 weeks after first defoliation), the root and rhizome systems of all plants were washed for the purpose of measurement. After rhizomes were severed from tap roots, each component was dried at 60°C for 48 h to determine the dry matter of roots and rhizomes.

Analysis of variance (ANOVA) was conducted to test the effects of all treatments on the traits measured using the Statistical Analysis System (SAS Institute Inc., 1998). Defoliation intensity, defoliation frequency, and kura clover entry were considered fixed, and replicates were considered random effects. To investigate the combined effect of defoliation intensity and frequency, another variable (DIF; defoliation intensity and frequency) was created to represent the combinations of values of defoliation intensity and defoliation frequency (Table 2). The combined effect (DIF) means were analyzed through a two-way classification using DIF and kura clover entry treatments. Fisher’s protected least significant difference (LSD) procedure was used to compare the treatment means.

Table 3. Root rhizome, and leaf production of kura clover entry, defoliation intensity, and defoliation frequency treatments in Exp. 1 (2002) and Exp. 2 (2003), Values are averaged across all other treatments

Treatment	Exp. 1			Exp. 2		
	Root	Rhizome	Leaf	Root	Rhizome	Leaf
	Dry matter (g)/plant					
Entry (ENT)						
R 1	2.87[c, 1]	1.05[a]	3.81[b]	2.24[c]	0.40[a]	2.13[b]
R 2	2.29[d]	0.25[b]	2.92[c]	2.62[c]	0.20[b]	2.10[b]
ARS-2678	4.77[a]	0.21[b]	4.44[a]	4.21[a]	0.08[b]	3.92[a]
Rhizo	3.62[b]	0.11[b]	3.58[b]	3.47[b]	0.16[b]	3.40[a]
Intensity (INT)[2]						
CD	2.87[b]	0.32[a]	3.46[b]	2.38[b]	0.10[b]	2.12[b]
PD	3.91[a]	0.48[a]	3.91[a]	3.89[a]	0.32[a]	3.65[a]
Frequency (FRE)[3]						
2-wk	1.44[c]	0.11[b]	2.70[b]	1.45[c]	0.06[b]	0.81[c]
4-wk	3.63[b]	0.39[b]	4.00[a]	3.09[b]	0.22[ab]	3.10[b]
6-wk	5.09[a]	0.71[a]	4.36[a]	4.85[a]	0.36[a]	3.74[a]
Contrasts and computed differences[4]						
Clone vs. population	-3.23*	0.98*	-1.36*	-2.82*	0.36*	-3.08*
Interaction						
ENT×INT	NS	NS	NS	*	NS	NS
ENT×FRE	*	NS	NS	*	NS	*
INT×FRE	NS	NS	*	NS	NS	NS
ENT×INT×FRE	NS	NS	NS	NS	NS	NS
	Pearson correlations coefficient					
Root vs. rhizome		0.18			0.31	
Root vs. leaf		0.83			0.92	

* Significant difference at the 0.05 probability levels.

[1] Means within a column followed by the same letter, within main effect, are not significantly different at 0.05 level of probability using a Fisher's LSD test.

[2] Defoliation intensity; CD means complete defoliation, PD means partial defoliation.

[3] Defoliation frequency; 2-wk means cut biweekly, 4-wk means cut every 4 weeks and 6-wk means cut every 6 weeks.

[4] First listed variable is positive and the second is negative in computation of differences.

RESULTS AND DISCUSSION

The root, rhizome, and leaf dry matter (DM) of four kura clover entries are shown in Table 3. In Exp. 1 and Exp. 2, ARS-2678 had excellent root development characteristics, however, its rhizome DM was significantly lower than R 1. Rhizo clones (R 1 and R 2) had lower root DM than ARS-2678 and Rhizo populations, however, R 1 had the greatest rhizome growth in Exp. 1 and Exp. 2. The leaf DM from these populations was significantly higher than that from Rhizo clones.

These results suggest that while root development was not directly related to rhizome development, root development showed a tendency to be associated with leaf development. The rhizome development from clones was faster than that from populations but the clones were developed from plants with known excellent rhizome growth. This result is consistent with the study of Speer and Allinson (1985) in which rhizome development was faster in plants propagated from rhizome segments than in plants from seed. However, root growth was greater in populations.

In Exp. 1 and Exp. 2, root and leaf DM production were affected by defoliation intensity (Table 3). The root and leaf DM under complete defoliation (CD) were significantly lower than under partial defoliation (PD). In Exp. 1, rhizome DM was not significantly different between CD and PD, but was significantly lower under CD in Exp. 2. King et al. (1978) reported a similar response in their white clover defoliation study. They found greater internode length of the stolon under partial versus complete defoliation conditions. Bell and Ritchie (1989) reported that a 3-cm defoliation height caused a greater reduction in root growth in prairie grass compared to a 8-cm defoliation height.

In Exp. 1 and Exp. 2, root, rhizome, and leaf DM generally increased with less frequent defoliation (Table 3). However, the increase in rhizome DM was not significantly different between 4- or 6-wk defoliation periods in either experiment. This result is similar to the results compiled by other researchers (Turner, 1968; Saldivar et al., 1992). Saldivar et al. (1992) compared the 2-, 6-, and 8-wk defoliation treatments with an undefoliated control in the rhizome production of 'Florigraze' rhizome peanut (*Arachis glabrata* Benth.).

Table 4. Dry matter (DM) and percentage of root, rhizome, and leaf production of defoliation intensity and frequency treatments (DIF) in Exp. 1 (2002) and Exp. 2 (2003), Values are means of 4 entries

Treatment[1]	Experiment 1						Experiment 2					
	Root		Rhizome		Leaf		Root		Rhizome		Leaf	
	DM (g)	%[2]	DM (g)	%	DM (g)	%	DM (g)	%	DM (g)	%	DM (g)	%
DIF[3]												
2-wk CD	0.85e[4]	16	0.08^{c}	9	2.23^{d}	52	0.98^{d}	17	0.01^{c}	2	1.14^{d}	25
4-wk CD	3.00^{c}	55	0.37^{bc}	41	3.70^{bc}	87	2.33^{c}	40	0.16^{bc}	27	2.22^{c}	50
6-wk CD	4.75^{b}	87	0.52^{ab}	58	4.67^{a}	110	3.83^{b}	65	0.12^{bc}	20	3.00^{b}	67
2-wk PD	2.04^{d}	38	0.14^{bc}	16	3.17^{c}	74	1.93^{c}	33	0.11^{bc}	19	2.48^{bc}	55
4-wk PD	4.26^{b}	78	0.41^{bc}	46	4.31^{a}	101	3.85^{b}	65	0.28^{b}	47	3.98^{a}	89
6-wk PD	5.43^{a}	100	0.90^{a}	100	4.26^{ab}	100	5.88^{a}	100	0.59^{a}	100	4.48^{a}	100
Interaction												
Entry[5]×DIF	*		NS		NS		*		NS		NS	

* Significance at the 0.05 levels of probability.

[1] Refer to Table 1 for description of treatment.

[2] Values are based on a 6-wk PD as being at 100% to clarify the difference among levels of DIF treatment.

[3] A created variable whose values are the combinations of values of defoliation frequency and intensity.

[4] Means within a column followed by the same letter, within main effect, are not significantly different at 0.05 level of probability using a Fisher's LSD test.

[5] R 1, R 2, ARS-2678 and Rhizo.

In the 2- and 4-wk defoliation frequencies in this study, fewer rhizomes developed. In the 8-wk defoliation frequencies, rhizome production decreased to about one-half that of the undefoliated control. Turner (1968) found that the rhizome weight of couch grass (*Agropyron repens* L. Beauv.) was consistently higher under infrequent defoliation (28-day) compared with frequent defoliation (14-day).

The intensity (INT)×frequency (FRE) of defoliation interaction was not significant ($p>0.05$) for root and rhizome DM in either experiment. The below-ground biomass was consistently greater under partial than complete defoliation, and it increased as defoliation intervals lengthened. Some ENT×INT or ENT×FRE interaction ($p<0.05$) occurred in root and leaf DM. This result indicates that root development of each entry did not show the same pattern under defoliation intensity or frequency treatments. However, entry treatments were averaged to clarify the effect of defoliation intensity and frequency (DIF), because ENT×INT or ENT×FRE interactions were usually a result of magnitude change and the focus of this study is defoliation severity (Table 4). ENT×INT or ENT×FRE interactions ($p>0.05$) for rhizome DM were not observed.

In both experiments, root DM was the greatest under a 6-wk PD treatment, next greatest under a 6-wk CD and 4-wk PD treatment, and the least under a 2-wk CD treatment (Table 4). Rhizome DM was superior under a 6-wk PD treatment, being over 10 and 50 times greater than under a 2-wk CD treatment, in Exp. 1 and Exp. 2, respectively. In Exp. 1, rhizome DM was not significantly different in all treatments except for a 6-wk CD and a 6-wk PD. In Exp. 2, in all treatment except for a 4-wk PD and a 6-wk PD, rhizome DM was not significantly different. Leaf DM was slightly (10%) greater, but not significantly higher in a 6-wk CD than a 6-wk PD, and was lowest in a 2-wk CD in Exp. 1. The leaf development was the greatest in a 4-wk PD and a 6-wk PD and least in a 2-wk CD in Exp. 2.

Overall, defoliation intensity and frequency treatments had an effect on root and leaf development to a much greater degree than on rhizome development. This result seems to be in part due to kura clover's slow initial growth. A 5-month period after planting appeared to be too short for kura clover to slow its rhizome growth characteristics relative to defoliation treatments. Root and leaf developments were usually the greatest to least as follow: in a 6-wk PD, a 6-wk CD or a 4-wk PD, a 4-wk CD or a 2-wk PD, and a 2-wk CD. These results were observed both in Exp. 1 and Exp. 2. The rhizome development was greatest in a 6-wk PD comparing production under all defoliation treatments.

In the present study, it was revealed that the intensity and frequency of defoliation should be minimized or defoliation should be avoided if maximum root and rhizome growth are desired from young kura clover plant. This study also imply the grazing should be delayed until the development of rhizome fully initiates, especially the heavy grazing should be inhibited in the establishment year for persistence of kura clover pasture.

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