

Regrowth capacity in relation to defence strategy in *Stipa clarazii* and *Stipa trichotoma*, native to semiarid Argentina

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Abstract Plants can defend themselves against herbivores by either avoiding or tolerating herbivory. Since avoidance mechanisms divert resources to other than growth processes, it could be expected a lower regrowth capacity in species that avoid herbivory than in species that tolerate herbivory, particularly under competition for resources. We tested this hypothesis by quantifying the regrowth of a grazing-avoidant (*Stipa trichotoma* Nees, synonymous *Nasella trichotoma* (Nees) Hackel ex Arechav.) and a grazing-tolerant (*Stipa clarazii* Ball, synonymous *Nasella clarazii* (Ball) Barkworth) grass species native to semiarid Argentina, when growing either singly or in pairs (one individual of each species) under repeated defoliation in field conditions. Twenty pairs of plants were selected in an area codominated by both species. Plants were protected by a 60 cm-diameter enclosure, in which the rest of the vegetation had been removed. The same procedure was followed with 20 single plants of each species. The same plants were clipped four times in 1996 and 1998 and five times in 1997. The response variable was the cumulative regrowth per plant at the end of each year. The regrowth was analysed for neutral detergent fibre and nitrogen content. Cumulative regrowth of *S. clarazii* was higher, similar, or lower than the cumulative regrowth of *S. trichotoma* in 1996, 1997, and 1998, respectively. Regrowth was reduced by interspecific competition, although there was no interaction between species and growing conditions (single or in pairs). Neutral detergent fibre content was consistently higher, whereas nitrogen content was consistently lower, in *S. trichotoma* than in *S. clarazii*. Collectively, our results did not support the hypothesis of higher regrowth capacity in the grazing-tolerant species than in the grazing-avoidant species. Further, our findings suggest herbivore tolerance is a previously unappreciated trait of *S. trichotoma*.

Key words: grazing avoidance, grazing tolerance, plant–herbivore interactions, *Stipa clarazii*, *Stipa trichotoma*.

INTRODUCTION

Plants can defend themselves against herbivores by either avoidance (a term used synonymously with avoidance is resistance) or tolerance of herbivory (Lindroth 1989; Briske 1991; Rosenthal & Kotanen 1994; Strauss & Agrawal 1999). Avoidance mechanisms reduce the probability and amount of defoliation, while tolerance mechanisms enhance a plant's ability to grow after defoliation. Examples of the former mechanisms are the production of secondary compounds, spines and thorns, leaf silicification, leaf sclerophylly, and basal meristematic tissues, whereas examples of the latter mechanisms are increased photosynthesis, tillering and carbon allocation from below-ground to above-ground tissues after defoliation. Ascertaining the original adaptive value of these mechanisms is difficult because of convergent selection pressures (Gould & Lewontin 1979). For example, adaptations to dry conditions (Coughenour 1985) and (or) low soil fertility (Rosenthal & Kotanen

1994) may also promote tolerance or avoidance of grazing.

It has been proposed that avoidance and tolerance mechanisms compete for resources generating a physiological trade-off, which results in a negative genetic correlation between avoidance and tolerance (e.g. Van der Meijden *et al.* 1988; Herms & Mattson 1992; Simms 1992). Thus, a genotype that invests more in avoidance has less resource available for tolerance and vice versa.

In semiarid grasslands of central Argentina both defence strategies (avoidance and tolerance) have been selected at the species level (Distel & Bóo 1996). Grasses can be clearly grouped into palatable species that tolerate grazing and unpalatable species that avoid grazing. Avoidance is mainly related to the development of sclerophyllous leaves (Distel *et al.* 2005), which are less expensive in terms of mineral nutrients (Beadle 1968) but more energetically expensive to construct than non-sclerophyllous leaves (Mooney & Gulmon 1982; Griffin 1994; Baruch & Gómez 1996). Sclerophylly may limit the regrowth capacity, particularly under competition for resources (Rosenthal & Kotanen 1994).

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The objective of our study was to quantify the regrowth of a grazing-avoidant (*Stipa trichotoma* Nees, synonymous *Nasella trichotoma* (Nees) Hackel ex Arechav.) and a grazing-tolerant (*Stipa clarazii* Ball, synonymous *Nasella clarazii* (Ball) Barkworth) grass species, when growing either singly or in pairs under recurrent defoliation in field conditions. In addition, the regrowth from both species was chemically analysed to help in the interpretation of the results.

METHODS

The study was conducted in the Caldén District (Cabrera 1976), on an upland site located in the south-eastern zone of La Pampa province in Argentina (38°45'S; 63°45'W). The site comprises a total area of 20 ha, from which livestock grazing has been excluded since 1980. The climate of the region is temperate and semiarid. Mean monthly temperatures range from a low of 7°C in July to a high of 24°C in January, with an annual mean of 15°C. Mean annual rainfall is 400 mm, with peaks in autumn and spring. The more severe droughts occur in summer. Rainfall in the experimental period was 530 mm, 548 mm and 375 mm, in 1996, 1997 and 1998, respectively. The monthly distribution of precipitation was normal over the 3 years of the study, except for the scarcity of rainfall at the end of winter to the beginning of spring of 1998. Dominant soils are Calciustolls, of coarse texture. A petrocalcic horizon is commonly found at depths of 60–80 cm.

The potential physiognomy of the vegetation is grassland with isolated woody plants (Distel & Bóo 1996). The more abundant herbaceous species are perennial cool-season bunchgrasses (family Poaceae), which carry out their annual growing cycle from March throughout December. Late seral dominants are mainly represented by the palatable midgrasses *S. clarazii* and *Poa ligularis*, whereas the palatable shortgrasses *S. tenuis* and *Piptochaetium napostaense* are mid-seral dominants. In uplands with a long history of heavy grazing the occurrence of patches dominated by unpalatable midgrasses is common. The more important species in this group are *S. gynerioides*, *S. tenuissima*, *S. trichotoma*, *S. speciosa* and *S. ambigua*. The palatable grasses are higher in protein and lower in structural carbohydrates than the unpalatable grasses (Cerqueira *et al.* 2004; Distel *et al.* 2005).

In March 1996 20 pairs of plants were selected in an area codominated by *S. trichotoma* (grazing-avoidant) and *S. clarazii* (grazing-tolerant). Pairs were composed of one individual of each species which were growing close together (with touching canopy). Plants were protected by a small enclosure (60 cm diameter by 40 cm tall) of reticulate mesh wire (5 cm by 5 cm). The rest of the vegetation within the enclosure had

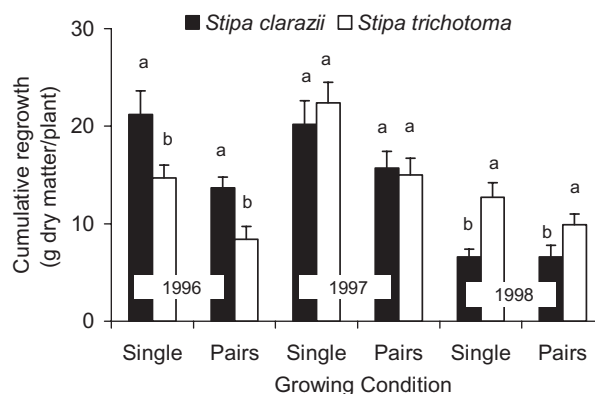


Fig. 1. Cumulative regrowth from four clippings (1996, 1998) or five clippings (1997), in plants of *Stipa clarazii* and *Stipa trichotoma* growing singly or in pairs (one plant of each species). Values are means ($n = 20$) \pm 1 standard error. Within year and growing conditions, different letters indicate a significant ($P < 0.01$) difference between means.

been carefully removed. The same procedure was followed with 20 single plants of each species. The average basal area per plant was 31.2 ± 3 (SE) cm^2 and 30.3 ± 3 (SE) cm^2 for *S. trichotoma* and *S. clarazii*, respectively. All plants were located within an area of approximately 0.5 ha, comprising separate vegetation patches. Immediately after enclosures were established, the above-ground biomass of each plant was clipped at 3 cm above the soil surface, to remove old standing dead biomass. Subsequently plants were clipped each time the regrowth in at least half of the individuals of one species reached 15–20 cm tall. The same plants were clipped four times in 1996 and 1998 and five times in 1997. The harvested material was dried at 60°C to a constant weight and weighed. The response variable in the analysis was the cumulative regrowth per plant at the end of each year.

The biomass from five plants of each species growing singly and five plants growing in pairs was analysed seasonally for neutral detergent fibre (Goering & Van Soest 1970) and nitrogen (semimicro Kjeldahl; AOAC 1980) content. The same plants were analysed throughout the experimental period. Plants were selected by using a list of random numbers.

Data were subjected to repeated measures analysis according to a completely random design. Factors were species, growing conditions (plants growing singly or in pairs), and time (years for cumulative regrowth or years and seasons for chemical parameters). The error term to test for species effect was individual plants nested within species.

RESULTS

Figure 1 shows the cumulative regrowth per plant of both species, growing either singly or in pairs, for each

Table 1. ANOVA assessing the effect of species, year and growing conditions (plants growing singly or in pairs) on cumulative regrowth of *Stipa clarazii* and *Stipa trichotoma*

Source	Mean square	d.f.	F	P
Species	13.27	1	0.14	0.7119
Year	1712.93	2	37.33	<0.0001
Growing conditions	1734.15	1	37.79	<0.0001
Species × year	550.74	2	12.00	<0.0001
Species × growing conditions	34.43	1	0.75	0.3875

d.f., degree of freedom; F, F-test.

Table 2. Neutral detergent fibre and nitrogen content of *Stipa clarazii* and *Stipa trichotoma*

Species	1996			1997			1998		
	Autumn, %	Winter, %	Spring, %	Autumn, %	Winter, %	Spring, %	Autumn, %	Winter, %	Spring, %
Neutral detergent fibre									
<i>S. clarazii</i>	59.0 (0.26)	60.4 (0.53)	62.4 (0.48)	60.5 (0.68)	61.8 (0.40)	64.5 (0.41)	57.0 (0.63)	61.6 (0.47)	63.9 (0.41)
<i>S. trichotoma</i>	61.5 (0.25)	62.4 (0.26)	72.0 (0.46)	70.0 (0.46)	71.3 (0.46)	73.9 (0.56)	60.8 (0.57)	64.2 (0.64)	70.3 (0.75)
Nitrogen									
<i>S. clarazii</i>	2.51 (0.09)	2.54 (0.04)	2.00 (0.03)	2.23 (0.09)	2.32 (0.08)	1.62 (0.05)	2.36 (0.06)	2.17 (0.07)	1.73 (0.04)
<i>S. trichotoma</i>	2.01 (0.05)	1.95 (0.05)	1.55 (0.05)	1.98 (0.06)	1.80 (0.06)	1.27 (0.06)	1.72 (0.04)	1.33 (0.05)	1.06 (0.04)

Values are average ($n = 10$) and 1 standard error. For each chemical parameter, all the differences between means within the year and season are significant ($P < 0.01$).

Table 3. ANOVA assessing the effect of species, season and year on neutral detergent fibre and nitrogen content of *Stipa clarazii* and *Stipa trichotoma*

Source	Neutral detergent fibre				Nitrogen			
	Mean square	d.f.	F	P	Mean square	d.f.	F	P
Species	1669.45	1	491.66	<0.0001	12.80	1	192.11	<0.0001
Season	641.81	2	173.65	<0.0001	5.98	2	176.09	<0.0001
Year	317.54	2	85.92	<0.0001	2.04	2	60.16	<0.0001
Species × season	62.49	2	16.91	<0.0001	0.15	2	4.48	0.0128
Species × year	116.88	2	31.62	<0.0001	0.44	2	13.03	<0.0001

d.f., degree of freedom; F, F-test.

year of measurement. Species did not differ in regrowth capacity ($P = 0.7119$), although the interaction species by time was significant ($P < 0.0001$) (Table 1). The cumulative regrowth of *S. clarazii* was higher, similar, or lower than the cumulative regrowth of *S. trichotoma* in 1996, 1997, and 1998, respectively. Regrowth was reduced ($P < 0.0001$) by interspecific competition (plants growing in pairs), although the interaction species by growing conditions (singly or in pairs) was not significant ($P = 0.3875$) (Table 1).

Since the growing conditions had no effect on chemical composition, data on neutral detergent fibre and nitrogen content were pooled across growing conditions (Table 2). Both chemical parameters were influenced ($P < 0.0001$) by species, season and year. Although the interactions species by season and species by year were significant (Table 3), the differences between species were always significant ($P < 0.01$). Fibre content was higher in *S. trichotoma* than in *S. clarazii* and it increased as seasons progressed, whereas nitrogen content was lower in the former than

in the latter species and it decreased as seasons progressed. For both species overall mean values of fibre content were higher ($P < 0.01$) in 1997 than in 1996 and 1998, whereas overall mean values of nitrogen content showed a decreasing ($P < 0.01$) trend through the years.

DISCUSSION

Our results did not support the hypothesis of reduced regrowth capacity associated with grazing avoidance. The cumulative regrowth of *S. trichotoma* (grazing-avoidant) was lower than that of *S. clarazii* (grazing-tolerant) in the first year of measurement, but it was similar or higher in the second and third year of measurement (Fig. 1). The trend in cumulative regrowth showed by *S. clarazii* paralleled the trend in precipitation, which was above the long-term average in 1996 and 1997 and below the long-term average in 1998, when precipitations were particularly scarce at the end of winter and the beginning of spring. The reduced regrowth observed for this species in 1998 may have been partially due to dry conditions. Repeated clipping events may have also progressively weakened the plants of *S. clarazii*. This species is characterized by large tillers, but in low numbers (low tiller density) (Distel & Klich 1996). Low tiller density translates into less remnant leaf area, which is critical for regrowth after defoliation (Briske 1991).

The cumulative regrowth of *S. trichotoma* followed a different trend. It was relatively low in 1996 and 1998 and highest in 1997 (Fig. 1). This trend appears to reflect a time lag for the expression of tolerance mechanisms in response to multiple defoliation events. Although a time lag has been reported for inducible chemical defences (Zangerl 2003), we were unable to find evidence for a lag to occur for tolerance mechanisms. On the other hand, the higher cumulative regrowth in *S. trichotoma* than in *S. clarazii* in 1998 may be attributed to better drought-tolerance, conferred in part by sclerophyllous leaves. Unfortunately, there are no data in our study on other plant properties that contribute to drought-tolerance. Alternatively, higher regrowth may have been due to larger remnant leaf area after defoliation due to higher tiller density in *S. trichotoma*. Tiller density can be up to four times higher in *S. trichotoma* than in *S. clarazii* (Andrea Flemmer, unpubl. data, 2001). The latter characteristic (high tiller density) may also contribute to explain the high regrowth capacity of *S. trichotoma*, even though their leaves are higher in neutral detergent fibre content and lower in nitrogen content than leaves of *S. clarazii* (Table 2). Relative growth rates at the tiller level is higher in *S. clarazii* than in *S. trichotoma*, but at the plant level it is higher in the latter than in the former species because of higher tiller density (Andrea

Flemmer, unpubl. data, 2001). Collectively, the results suggest that *S. trichotoma* has a relatively high capacity to tolerate defoliations. Therefore, both defence strategies, avoidance and tolerance, appear to have been selected in this species. Our results did not support the hypothesis that regrowth capacity after defoliation is higher in palatable than in unpalatable grasses (Noy-Meir & Walker 1986). In contrast, the hypothesis was supported in a comparison between the unpalatable grass *Aristida ramosa* and the palatable grass *Danthonia linkii* in Australia (Harradine & Whalley 1981; Lodge & Whalley 1985).

In a recent review on the evolutionary ecology of tolerance to consumer damage, Stowe *et al.* (2000) argued that there are species where selection has favoured the retention of both avoidance and tolerance mechanisms. This is consistent with the absence of a negative genetic correlation between avoidance and tolerance (Mauricio *et al.* 1997). de Jong and van der Meijden (2000) argued that if a genotype has evolved under conditions of multiple sources of damage, an avoidance-tolerance trade-off should not be expected. There are abiotic disturbances such as drought that can not be avoided. Since drought is a recurrent disturbance in the system where *S. trichotoma* has evolved, and because damaged leaves have to grow quickly and expand to full-size before the woody materials characterising the hard sclerophyllous leaves can be deposited, this species appears to have evolved both tolerance and avoidance mechanisms. Further, since the system has evolved under low pressure from large mammalian herbivores (Webb 1978; Bucher 1987), avoidance mechanisms present in *S. trichotoma* and other unpalatable species (e.g. *S. tenuissima*, *S. gynerioides*) may have arisen as adaptations to survive drought and are incidentally beneficial to avoid grazing (exaptations, *sensu* Coughenour 1985). Alternatively, they might have evolved as adaptations to conditions of low soil fertility (Rosenthal & Kotanen 1994), which are common in the study area (INTA *et al.* 1980). For example, sclerophylly (high fibre content) is a common evolved plant property on soils of low fertility in the semiarid and arid zone of Australia (Beadle 1968; Harradine & Whalley 1978; Lodge & Whalley 1983), a country where the grasslands apparently also evolved under low grazing pressure (Donald 1970; Suijendorp 1980).

Our results may contribute to explain the remarkable persistence of some unpalatable grasses once they attain a high level of dominance in areas formerly dominated by palatable grasses (Noy-Meir & Walker 1986). It has been argued that low frequency and high intensity of grazing, by reducing herbivore selectivity, may enable palatable species to recover dominance (Anderson & Briske 1995; Moretto & Distel 1999). However, our findings suggest herbivore tolerance is a previously unappreciated trait of some unpalatable

grasses that, in conjunction with avoidance traits, are likely to contribute to their persistence.

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