

Nutrient acquisition and physiological responses of dominant Serengeti grasses to variation in soil texture and grazing

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Summary

1 *Themeda triandra* and *Digitaria macroblephara* are dominant grasses in the herbaceous layer of Serengeti National Park. Catena topography creates variation in soil texture and grazing that may have important physiological implications for these species. Through a combination of laboratory and field experiments, we tested whether variation in soil texture and grazing differentially affected growth, photosynthesis, leaf enzyme activities and nitrogen assimilation of the two species.

2 In the laboratory, simulated grazing (weekly clipping at 7 cm above soil) increased photosynthesis and enzyme activities for *D. macroblephara*. Clipping also increased photosynthesis for *T. triandra*, but did not affect enzyme activity. *D. macroblephara* and *T. triandra* were affected oppositely by soil texture; enzyme activities and photosynthesis were greater for *D. macroblephara* in sandy soils but greater for *T. triandra* in fine-textured soils.

3 Across a natural catena gradient, species abundances of *D. macroblephara* and *T. triandra* were negatively correlated with each other and related to grazing frequency in opposite ways, *D. macroblephara* positively and *T. triandra* negatively. In a field experiment, rates of $^{15}\text{NO}_3^-$ uptake for unclipped plants of *D. macroblephara* and *T. triandra* were similar and did not vary with soil texture. However, when clipped, $^{15}\text{NO}_3^-$ uptake was stimulated in both species but in opposite ways across a gradient of percentage sand, positive for *D. macroblephara* and negative for *T. triandra*.

4 These results suggest that species responded to defoliation and variation in water availability created by different soil textures. At sites where *D. macroblephara* and *T. triandra* co-occur, catena variation in soil texture and grazing may provide spatial variation in habitat suitability. Moreover, herbivory may promote species coexistence by facilitating differential physiological responses to topographic and soil heterogeneity.

5 This study illustrates how interactions between a direct biotic process and an indirect environmental variable can cause different plant physiological responses that may promote species coexistence in a heterogeneous environment.

Key-words: ^{15}N uptake, catena position, defoliation, environmental heterogeneity, enzyme activity, photosynthesis, soil texture, species coexistence, topographic variation

Journal of Ecology (2006) **94**, 1164–1175

doi: 10.1111/j.1365-2745.2006.01148.x

Introduction

For many herbaceous plants, soil heterogeneity provides a physical template to which species have different

germination success, growth capabilities, reproductive capacity and competitive abilities (Davies *et al.* 1998; Day *et al.* 2003; Wijesinghe *et al.* 2005). Soil texture is an important indirect environmental variable (Ausin 1980; Ausin & Smith 1989) that influences plants through its effects on microbial dynamics and water and nutrient availability (Schimel *et al.* 1985; Groffman *et al.* 1996; Hook & Burke 2000; Barrett & Burke 2002). Likewise, defoliation by herbivores, a direct biotic process, affects

the growth (Ferraro & Oesterheld 2002), reproduction (Anderson & Frank 2003) and competitive abilities (Berendse 1985; van der Wal *et al.* 2000) of herbaceous species. There is reason to believe that soil texture and defoliation interact because the effects of herbivores on plant production depend on water and nutrient availability (Georgiadis *et al.* 1989; Wise & Abrahamson 2005). Interactions between grazing and soil affect plant community patterns (Burke *et al.* 1999; Stohlgren *et al.* 1999), but less known is how the physiological responses of plants to interactions between soil and herbivory translate into different habitat affinities in nature.

In the Serengeti National Park (SNP), Tanzania, over 350 plant species coexist in the grassland plant communities (Anderson 2004), but grasses dominate the biomass of the herbaceous layer (McNaughton 1985). Multiple environmental factors influence plant community patterns and coexistence, in savannas typical of the Serengeti, including fire (McNaughton 1983), nutrients (Ludwig *et al.* 2001; Anderson *et al.* 2004), rainfall (McNaughton 1985), soil factors, such as texture and type (Anderson & Talbot 1965; de Wit 1978; Jager 1982; Belsky 1986), and grazing herbivores (McNaughton 1983, 1985). Several authors have suggested that variability in SNP catena topography creates variations in soil texture and water availability that affects plant species coexistence at these spatial scales, from dry, sandy hilltops to the moist, heavy clay sumps found in the bottom hillslopes (Bell 1970; de Wit 1978; Jager 1982). These anecdotal hypotheses have suggested that soil microsite conditions are responsible for the distribution of 'long grass species' in bottom hillslopes and 'short grass species' on upper hillslopes (Bell 1970), thus promoting coexistence at these relatively small scales. However, grazing also varies at these spatial scales and shifts along hillslope positions, as grazers utilize the wetter, bottom hillslope positions in the dry season and drier hilltops in the wet season (Bell 1971; Seagle & McNaughton 1992). Therefore, grazing and soil conditions may work interactively to affect the habitat conditions of different Serengeti grasses.

Two C_4 perennial bunch grasses, *Digitaria macroblephara* and *Themeda triandra*, dominate SNP grasslands in terms of cover, biomass and frequency (Anderson 2004). *D. macroblephara* is medium-stature, invests heavily in stolon growth, and was classified as a top-hillslope grass by Bell (1970). *T. triandra* is an obligate seed producer of taller stature and was classified as a bottom-hillslope grass (Bell 1970). In terms of resilience to herbivory, *D. macroblephara* is considered grazing-tolerant, while *T. triandra* is less tolerant to frequent defoliation (Heady 1966; Coughenour *et al.* 1985; Hodgkinson *et al.* 1989; O'Connor 1994; Fynn & O'Connor 2000). Observational evidence that soil and defoliation may differentially affect *D. macroblephara* and *T. triandra* was provided by a multivariate ordination of Serengeti plant communities, in which two factors, annual grazing intensity and soil texture, showed the greatest correlation with the distribution of 17 plant community types (McNaughton

1983). Plant communities dominated by *T. triandra* were associated with clay soils and low annual grazing intensities compared with *D. macroblephara* communities, which were associated with sandy soils and higher annual grazing intensity (McNaughton 1983).

Given the importance of soil texture and grazing to SNP plant communities and because these factors display spatial variation at scales relevant to the coexistence of dominant grasses, we hypothesized that soil texture and grazing would differentially affect *D. macroblephara* and *T. triandra* and may interact to promote species coexistence. While other factors such as fire and rainfall have important effects on grassland plant communities across the SNP environmental gradient, we choose to focus on the effects of soil texture and defoliation because (i) grazing and soil texture were identified in previous research as having the strongest association with Serengeti plant community composition (McNaughton 1983); and (ii) because these species have displayed inconsistent responses to other factors in this system, such as fire (Belsky 1983; McNaughton 1983; McNaughton 1985; Belsky 1992), suggesting that plant responses to defoliation due to fire may depend on other underlying factors.

In this study, we investigated the importance of soil texture and defoliation for the growth, physiology and nutrient acquisition of *T. triandra* and *D. macroblephara*. In the laboratory, we determined the effects of variation in soil texture and repeated defoliation on growth, leaf-level photosynthesis, enzyme activities and nitrogen (N) acquisition. Enzyme analyses focused on: pyruvate phosphodikinase (PPDK) and phosphoenolpyruvate carboxylase (PEPc), both C_4 photosynthesis enzymes, nitrate reductase (NR) and glutamine synthetase (GS), N assimilation enzymes. PPDK catalyses the first reaction of C_4 photosynthesis and is the rate-limiting enzyme (Usuda *et al.* 1984). PEPc catalyses the subsequent reaction accompanied by the highly efficient assimilation of CO_2 from atmosphere. NR catalyses the reduction of nitrate and GS catalyses the assimilation of ammonium, the final form of all N assimilated by plants. In a field experiment, we transplanted *T. triandra* and *D. macroblephara* into artificial plots to study how soil texture and defoliation affected N assimilation. We focused on N assimilation because it is indicative of plant fitness in an N-limited system such as the Serengeti (McNaughton *et al.* 1983; McNaughton *et al.* 1988) and it determines the concentration of other trace minerals in SNP grasses (Hamilton *et al.* 1998).

Methods

LABORATORY EXPERIMENT

The effects of soil texture and repeated clipping on leaf-level physiological processes and N uptake in *D. macroblephara* and *T. triandra* were measured in 8-week laboratory studies. Twenty individuals of each species were propagated at Syracuse University and grown in the glasshouse for 6 months. Subsequently, propagates

were reduced to approximately 10 tillers per plant and randomly assigned to one of four factorial treatment combinations of soil texture (high or low sand) and simulated weekly grazing (clipped and unclipped). Soils were prepared by mixing Agway topsoil (Scotts Company, Lebanon, CT) with course grain sand (Amstone, Towson, MD) in volumetric ratios of 3 : 1 and 2 : 3 soil to sand, resulting in treatments of 50% and 75% sand, respectively. The range of soil texture levels was representative of those in which *D. macroblephara* and *T. triandra* coexist in Serengeti grasslands (McNaughton 1983; Anderson *et al.* 2004), and were chosen to reflect the natural range of conditions that plants encounter across a typical SNP catena gradient (de Wit 1978; Jager 1982; McNaughton 1985). Plants were grown in pots made from 10 cm diameter, 30 cm tall PVC pipes sealed with silicone to a 10 × 10 cm ceramic tile with holes in the base to allow water drainage. Plants were clipped to 12 cm 1 week prior to the start of the experiment. Pots were randomly placed in the growth chamber and locations were randomized; clipped plants were elevated on wooden blocks to eliminate shading effects. Plants were grown on a 12 : 12 hour light : dark cycle following a unimodal cycle in 2-hour increments with the following photosynthetic active radiation (PAR) levels: 245, 445 and 575 $\mu\text{m}^{-2} \text{s}^{-1}$. PAR was only about 40% of typical midday light levels in the Serengeti (T. M. Anderson, personal observation), but was the maximum light level produced by the growth chamber. Because soil texture alters nutrient availability, and effects of nutrients and grazing on Serengeti grasses have been well studied (McNaughton *et al.* 1983; McNaughton & Chapin 1985; Ruess 1988; Hamilton *et al.* 1998), we elected to reduce the effects of nutrient deficiency as a possible mechanism responsible for observed treatment differences. To this end, plants were fertilized once a week with 120 mL Hoagland's solution No. 3 in addition to receiving 120 mL deionized water twice a week.

Weekly procedures, in the following order, consisted of: measurement of photosynthesis, clipping plants in the simulated grazing treatment, and fertilizing. Tissue collection for enzyme analysis was conducted biweekly. Photosynthesis, leaf internal CO_2 and stomatal conductance were measured in the growth chamber at peak light level with a Licor 6200 IRGA. Measurements were made on two mature, recently expanded leaves in the upper-canopy of each plant. Plants in the grazing treatment were clipped to 7 cm once a week, a height and frequency representative of grassland swards occupied by *D. macroblephara* and *T. triandra* in the mid-grass region of the Serengeti (McNaughton 1984, 1985). The same grazing height was used for both species because herbivores maintain SNP mid-grass swards at a uniform height for more than 6 months of the year (McNaughton 1984, 1985). Above-ground biomass removed during clipping was dried at 65 °C for 48 hours and weighed; above-ground weekly off-take was combined with total harvested biomass to obtain the final biomass of clipped plants. To ascertain the influence of soil texture and defoliation

on soil water, soil volumetric water content was measured weekly at a depth of 12 cm with a HydroSense® time domain reflectometer (Campbell Scientific, Logan, Utah, USA).

Plant N acquisition from soils of different texture was studied by injecting a K^{15}NO_3 solution into each pot 5 days before the end of the experiment, after which plant tissue was analysed for ^{15}N . The solution consisted of 0.666 mMol litre⁻¹ ^{15}N , prepared by mixing 0.063 g K^{15}NO_3 (99 atom percentage ^{15}N ; Aldrich Chemical Co., Milwaukee, WI, USA) litre⁻¹ water. A pilot study suggested that both $^{15}\text{NO}_3^-$ and $^{15}\text{NH}_4^+$ would perform equally well at demonstrating the N uptake dynamics of *D. macroblephara* and *T. triandra*, a finding consistent with that of other SNP grasses (Ruess 1988). Twelve 2-mL aliquots, spaced 2 cm apart with the plant in the centre, were injected in each pot to a depth of 10 cm with a modified four-port refillable veterinary syringe (McKane *et al.* 2002). Care was taken that no contact was made between the above-ground biomass and syringe. Injections delivered 0.004 g $^{15}\text{N} \text{m}^{-2}$, an amount estimated to increase isotopic signatures above 50‰, while minimizing plant fertilization (R. B. McKane, personal communication). Above- and below-ground plant tissues were harvested separately to avoid contamination. Tissues were dried in an oven at 65 °C for 48 hours and weighed. Above-ground plant tissues were homogenized in a Wiley mill and analysed for ^{15}N at UC Davis stable isotope facility.

LABORATORY PROTEIN AND ENZYME ANALYSIS

In the laboratory experiments, enzyme activities of PPK, PEPc, GS and NR were assayed in the leaves of *D. macroblephara* and *T. triandra* following 2, 4, 6 and 8 weeks. Approximately 0.2 g of leaf tissue was sampled from fully developed young leaves of each plant and stored at -80 °C until analysis. When processed, each sample was ground into a fine powder in liquid N and homogenized in 3 mL extracting buffer with a mortar and pestle as in Scheible *et al.* (1997). Immediately after extraction, PPK was assayed spectroscopically at 340 nm, as described by Ashton *et al.* (1990). Reactions began by adding 200 μL extracted sample to 800 μL reaction mixture and monitoring the absorbance decrease at 340 nm (NADH oxidation). PEPc activity was measured with a spectrophotometer as the rate of PEP-dependent NADH oxidation at 340 nm, as described in Scheible *et al.* (1997). GS activity was measured with a spectrophotometer as the rate of NADH oxidation at 340 nm, as described in Kingdon *et al.* (1968). The reaction began with the addition of 100 μL extracted sample to 900 μL reaction mixture. NR activity was measured according to Brewitz *et al.* (1996), with NaNO_2 used to produce a standard curve. Mean enzyme activities were calculated per unit dry weight by correcting for the mass of water evaporated from samples of fresh tissue collected at the time of sampling.

CATENA FIELD EXPERIMENT

Between 1 March and 1 June 2002, corresponding with the growing season, a randomized block field experiment was conducted to determine the effects of soil texture and simulated grazing on growth and N uptake of *D. macroblephara* and *T. triandra*.

The field experiment was designed to meet two main criteria. The first was to ensure that plants experienced a range of soil textures, as we would not know the actual values until samples were returned to the laboratory at Syracuse University. We met this objective by experimentally manipulating soil texture. Secondly, we aimed to control for unknown effects of catena position not associated with soil texture. Unaccounted for spatial variation may arise for many reasons, for example different catena positions may create differences in water runoff. This criterion was met by experimentally manipulating soil texture within blocks at different positions along a catena gradient. Five blocks were located on average, 350 m from each other on a gently sloping catena gradient near the Serengeti Wildlife Research Center (SWRC) in the central region of SNP. Blocks consisted of nine 1-m² subplots randomly assigned to either a (i) control, (ii) intermediate sand or (iii) high sand treatment. Subplots were spaced 0.5 m apart, cleared of vegetation, and homogenized to a depth of 20 cm. Sand was added to subplots by replacing one (intermediate sand) or two (high sand) 19-litre buckets of native soil with the same volume of pure sand. Soil texture was determined using the Buoycous method (Elliot *et al.* 1999) on 40-g subsamples collected from each subplot. Soil gravimetric water content (g H₂O 100 g soil⁻¹) was measured on 10-g subsamples collected to a depth of 10 cm that were weighed immediately after collection, transported to Syracuse University, and re-weighed after drying at 105 °C for 48 hours. We acknowledge that gravimetric water content is not synonymous with plant available water and serves only as a measure of total soil water content in our study.

Nine individuals of *D. macroblephara* and *T. triandra* were collected adjacent to each block, weighed with a portable balance, randomly paired with conspecifics (six pairs) or heterospecifics (three pairs) of similar weight, and transplanted into subplots. Two individuals were planted in each subplot located 20 cm apart from each other. To reduce variation among replicate plants, roots were clipped to a length of 15 cm before transplanting, and shoots were clipped to a height of 5 cm after transplanting. Plants were watered immediately with 250 mL and again 1 week after planting. Methods used to study plant N acquisition in the field were identical to that in the laboratory with the following exceptions. In the field subplots, 20 injections were spaced 5 cm apart in a 4 × 5 cm grid with the target plant in the centre. The injections delivered 0.008 g ¹⁵N m⁻², an amount estimated to increase isotopic signatures above 100‰, while minimizing plant fertilization (R. B. McKane, personal communication). Only plants from subplots

containing conspecific individuals were injected ($n = 60$), except when a plant from a subplot containing heterospecific plants was randomly chosen to replace a plant that had died during the experiment. Of the 60 injected plants, half were randomly selected as unclipped control plants; the remainder were clipped to a height of 10 cm immediately following injection. After 5 days, above- and below-ground plant tissue was harvested and analysed as previously described. Although plants in the field experiment were not protected from grazing for the 12 weeks prior to ¹⁵N injections, most defoliation occurred soon after transplanting; only seven plants showed evidence of defoliation in the final 3 weeks before the injections, and only one in the final week.

To establish the natural pattern of abundance of *D. macroblephara* and *T. triandra* across the catena gradient and to identify associations between species' abundance and soil texture and grazing frequency, observational samples were collected in the undisturbed grassland adjacent to the five blocks. *D. macroblephara* and *T. triandra* abundance were measured by clipping all above-ground biomass in six randomly placed 0.5-m² plots surrounding each block once in March and again in May ($n = 12$). Clippings were sorted to species, air dried, transported to Syracuse University, oven dried at 65 °C for 48 hours and averaged by species and block to estimate above-ground biomass. Soil texture and gravimetric water content of undisturbed soil was measured in March and May as previously described. Soils sampled in March and May represented a range of soil moisture conditions because of the absence of significant January and February rainfall and abundant rainfall in April and May. Finally, grazing frequency at each block was calculated as the proportion of transplanted individuals from the catena experiment with evidence of defoliation each week averaged over all sampling dates ($n = 9$).

STATISTICAL PROCEDURES

Treatment means and residual errors from the laboratory experiment were investigated for collinearity prior to statistical analyses. Residual errors were correlated among physiological measures (photosynthesis, leaf internal CO₂, and stomatal conductance) and among three of the enzyme activities (PPDK, PEPc and GS). Therefore, responses were analysed with 2 × 2 × 2 factorial MANOVA (species × clipping × sand). For NR enzyme activity, total, leaf and root laboratory biomass, soil volumetric water content and laboratory ¹⁵NO₃⁻ uptake, treatment means were analysed with 2 × 2 × 2 factorial ANOVA. Our results focus on significant two- and three-way interactions that involve a 'species' term because we were interested in whether dominant species respond differently to catena variation in soil texture and grazing. Means (± SE) from two-way interactions are reported when three-way interaction effects were not significant. Significance between specific contrasts were determined after accounting for the false discovery rate (FDR) at $\alpha = 0.05$. FDR corrects for error introduced

Table 1 Mean (\pm SE) treatment responses of *Digitaria macroblephara* and *Themeda triandra* to two levels of clipping (clipped or unclipped) and soil texture (high or low sand) in the laboratory. Means are reported from two-way (a) or three-way (b) interactions among species, clipping and soil texture; two-way interactions under headings in italics (*leaf physiology* and *enzyme activity*) represent multivariate treatment responses, while total biomass, leaf biomass, root biomass, soil water, NR and ^{15}N uptake are univariate responses (see text)

(a) Two-way interactions

Response	<i>D. macroblephara</i>		<i>T. triandra</i>		<i>D. macroblephara</i>		<i>T. triandra</i>	
	Clipped	Unclipped	Clipped	Unclipped	High sand	Low sand	High sand	Low sand
Biomass	–	–	–	–	12.1 \pm 0.6 ^a	11.7 \pm 1.5 ^a	9.1 \pm 1.1 ^b	11.8 \pm 1.7 ^a
Leaf	–	–	–	–	7.4 \pm 0.3 ^a	7.2 \pm 0.8 ^a	5.3 \pm 0.6 ^b	7.4 \pm 1.1 ^a
Root	1.4 \pm 0.1 ^c	2.1 \pm 0.2 ^b	1.3 \pm 0.2 ^c	3.0 \pm 0.4 ^a	–	–	–	–
Soil water	9.3 \pm 0.7 ^b	6.5 \pm 0.3 ^c	15.5 \pm 2.1 ^a	8.7 \pm 1.1 ^{bc}	–	–	–	–
<i>Leaf physiology</i>								
Leaf A_o	17.8 \pm 0.2 ^a	15.3 \pm 0.3 ^b	15.1 \pm 0.5 ^b	13.3 \pm 0.5 ^c	17.0 \pm 0.5 ^a	16.0 \pm 0.6 ^b	13.2 \pm 0.4 ^c	15.3 \pm 0.5 ^b
Leaf C_{int}	185.4 \pm 3.1 ^b	164.9 \pm 3.9 ^c	291.6 \pm 2.2 ^a	291.5 \pm 4.3 ^a	175.2 \pm 5.3 ^c	175.1 \pm 5.1 ^c	291.0 \pm 2.1 ^a	286.1 \pm 3.3 ^b
Leaf s	0.13 \pm 0.004 ^c	0.11 \pm 0.003 ^d	0.22 \pm 0.007 ^a	0.20 \pm 0.004 ^b	0.13 \pm 0.007 ^b	0.11 \pm 0.006 ^b	0.20 \pm 0.005 ^a	0.21 \pm 0.009 ^a
<i>Enzyme activity</i>								
PPDK	4.7 \pm 0.2 ^a	2.7 \pm 0.1 ^c	3.4 \pm 0.2 ^b	3.4 \pm 0.3 ^b	4.0 \pm 0.4 ^a	3.5 \pm 0.3 ^b	2.9 \pm 0.1 ^c	3.9 \pm 0.1 ^a
PEPc	283.3 \pm 12.9 ^a	176.8 \pm 3.1 ^c	198.4 \pm 10.8 ^{bc}	204.3 \pm 10.8 ^b	241.9 \pm 25.3 ^{ab}	218.2 \pm 17.6 ^{bc}	183.4 \pm 10.3 ^c	219.3 \pm 6.1 ^{bc}
GS	10.5 \pm 0.3 ^b	6.7 \pm 0.2 ^c	26.1 \pm 1.2 ^a	25.8 \pm 1.2 ^a	8.9 \pm 0.9 ^b	8.3 \pm 0.6 ^b	25.0 \pm 1.3 ^a	26.9 \pm 1.1 ^a

(b) Three-way interactions

Response	<i>D. macroblephara</i>				<i>T. triandra</i>			
	Clipped		Unclipped		Clipped		Unclipped	
	High sand	Low sand	High sand	Low sand	High sand	Low sand	High sand	Low sand
NR activity	66.9 \pm 0.8 ^a	63.3 \pm 5.9 ^a	37.1 \pm 2.3 ^c	30.0 \pm 1.5 ^c	42.8 \pm 3.7 ^{bc}	42.4 \pm 2.8 ^{bc}	35.5 \pm 2.1 ^c	50.1 \pm 2.6 ^b
^{15}N uptake	253.8 \pm 33.8 ^a	37.4 \pm 9.2 ^b	67.1 \pm 10.9 ^b	27.2 \pm 9.2 ^b	21.5 \pm 4.2 ^b	17.5 \pm 3.0 ^b	30.9 \pm 3.3 ^b	27.8 \pm 4.6 ^b

Abbreviations are: biomass = final total (above + below) biomass (g plant^{-1}); leaf = final above-ground biomass (g plant^{-1}); root = final root biomass (g plant^{-1}); soil water = soil volumetric water content (%); A_o = photosynthesis ($\mu\text{mol m}^{-2} \text{s}^{-1}$); C_{int} = leaf internal CO_2 (p.p.m.); s = stomatal conductance ($\mu\text{mol m}^{-2} \text{s}^{-1}$); PPDK = pyruvate phosphodikinase (activity units gram^{-1}); PEPc = phosphoenolpyruvate carboxylase (activity units gram^{-1}); GS = glutamine synthetase (activity units gram^{-1}); NR = nitrate reductase (activity units gram^{-1}); and ^{15}N uptake = acquisition of soil ^{15}N tracer ($\mu\text{g plant}^{-1}$). Means followed by same letter are not significantly different ($\alpha = 0.05$) by pairwise comparisons after adjusting for the false discovery rate (see text); two-way comparisons are made within but not between levels of clipping or soil texture.

by conducting multiple comparisons while maximizing detectable differences between treatments (Verhoeven *et al.* 2005). For all laboratory analyses, parametric assumptions were met so that transformations were not necessary.

In the catena field experiment, relationships among observational data were investigated with regression and Pearson's correlation coefficients. Associations between species abundance and variation in defoliation and soil texture were analysed with separate stepwise multiple regressions (α to enter = 0.25, α to stay = 0.10). Soil percentage sand, gravimetric water content, and grazing frequency at five catena locations, were used to predict above-ground biomass of *T. triandra* and *D. macroblephara* in replicate 0.5-m² plots along the catena gradient. Final biomass and gravimetric water content from the transplant experiment were analysed with a 2 \times 3 factorial ANOVA in a randomized complete block design, with species (two levels) and soil texture (three levels) as main effects. The effects of simulated grazing and soil texture on rates of ^{15}N uptake were tested with a 2 \times 2 factorial ANCOVA, with species and clipping as main

effects and percentage sand as a covariate. Specific contrasts among regression coefficients of ^{15}N uptake vs. percentage sand for different treatment combinations were analysed with *t*-tests for differences between slopes. SAS version 9.1 (SAS Institute 2003) was used for all statistics.

Results

LABORATORY EXPERIMENT

Biomass

There was suggestive evidence that the final total biomass of *D. macroblephara* and *T. triandra* depended on soil texture ($F_{1,24} = 3.51$, $P = 0.07$), but no evidence that it depended on clipping ($F_{1,24} = 1.47$, $P = 0.24$). The species-by-sand interaction was a result of *T. triandra* grown in high sand, which had a lower final biomass than the other three treatments (Table 1). Moreover, the lower total biomass of *T. triandra* in the high sand treatment could be attributed to lower above-ground biomass,

resulting in a species-by-sand interaction ($F_{1,24} = 6.71$, $P = 0.016$; Table 1). There was little evidence that differences in above-ground biomass of the species depended on clipping ($F_{1,24} = 2.78$, $P = 0.11$). Root biomass displayed a significant species-by-clipping interaction ($F_{1,24} = 6.46$, $P = 0.018$), in which unclipped *T. triandra* had the highest root biomass, followed by unclipped *D. macaroblephara*, followed by clipped plants of which neither species differed (Table 1).

Photosynthesis

In terms of multivariate physiological responses (Table 1), the response of species depended on both clipping (Wilk's $\lambda = 0.56$, $F_{3,23} = 6.05$, $P = 0.0034$) and soil texture (Wilk's $\lambda = 0.39$, $F_{3,23} = 11.96$, $P < 0.0001$). For *D. macrolephara*, clipping increased photosynthesis, internal leaf CO_2 and stomatal conductance. For *T. triandra*, clipping also increased photosynthesis and stomatal conductance but with no associated change in internal leaf CO_2 . Rates of photosynthesis in clipped *D. macrolephara* were greater than for *T. triandra*, whereas unclipped *D. macrolephara* were similar to clipped *T. triandra*. Unclipped *T. triandra* had the lowest rates of photosynthesis. In contrast, internal leaf CO_2 and stomatal conductance were greater in *T. triandra* than *D. macrolephara* regardless of clipping treatment. In response to soil texture, the two species had opposite responses: *D. macrolephara* had the greatest rates of photosynthesis in high sand, while *T. triandra* had the greatest photosynthetic rates in low sand. However, photosynthesis of *D. macrolephara* and *T. triandra* in low sand was equivalent, while high sand had a strong negative effect on *T. triandra* photosynthetic rate. Stomatal conductance and internal leaf CO_2 in *D. macrolephara* were unaffected by sand and both were lower than *T. triandra* regardless of soil texture. Soil texture had no discernable effect on stomatal conductance in *T. triandra*, but those plants grown in high sand had greater leaf internal CO_2 than those in low sand.

Enzyme activities

In terms of multivariate response of enzyme activities (Table 1), the response of species depended on both clipping (Wilk's $\lambda = 0.16$, $F_{3,23} = 39.23$, $P < 0.0001$) and on soil texture (Wilk's $\lambda = 0.25$, $F_{3,23} = 21.96$, $P < 0.0001$). Clipping had no effect on PPDK activity in *T. triandra*, but increased PPDK activity in *D. macrolephara* by a factor of 1.74. PPDK activities of the two species were opposite in response to soil texture. *D. macrolephara* grown in high sand and *T. triandra* in low sand, had similar PPDK activities, while low sand inhibited *D. macrolephara* and high sand inhibited *T. triandra* PPDK activity. Responses of PEPc to the treatments were more complex, but in general clipping increased PEPc activity by a factor of 1.6 and when grown in high sand, *D. macrolephara* had greater PEPc activities than *T. triandra* grown in high sand. Univariate comparisons

did not detect significant differences between *T. triandra* grown in different soil textures or exposed to different defoliation treatments. For GS, *T. triandra* had over twice the activity levels of *D. macrolephara*, but showed no response to either clipping or soil texture. For *D. macrolephara*, GS activity did not respond to variation in soil texture but was stimulated by defoliation.

In contrast to other enzymes, NR was analysed with univariate statistics because of uncorrelated residual errors. NR activity showed a three-way interaction among species, clipping and soil texture ($F_{1,24} = 4.45$, $P = 0.04$). Readers are referred to Table 1 rather than discussing the 28 potential contrasts; we focus here on those results in which species deviate in NR enzyme activity under different soil and grazing conditions. For *D. macrolephara*, clipped plants grown in high and low sand had the highest NR enzyme activities, which were greater than the NR activities of *T. triandra* under the same conditions. In contrast, for *T. triandra*, unclipped plants grown in low sand had the highest mean NR activity, which was significantly greater than the NR activity level of unclipped *D. macrolephara* grown in low sand.

^{15}N uptake

After a single injection of K^{15}NO_3 at the end of the laboratory experiment, plant assimilation of $^{15}\text{NO}_3^-$ displayed a three-way interaction among species, clipping and soil texture ($F_{1,24} = 20.74$, $P = 0.0001$). However, the effect was attributable solely to the response of clipped *D. macrolephara* grown in high sand, which was more than three and a half times more than unclipped *D. macrolephara* grown in high sand, which was the second highest treatment (Table 1). Although some variation existed among the remaining treatment means, the relatively large discrepancy between clipped *D. macrolephara* grown in high sand and other treatments reduced the ability of univariate contrasts to detect differences. The large increase in $^{15}\text{NO}_3^-$ uptake by *D. macrolephara* could not be attributed to differences in size, as neither total biomass ($\rho_{15} = -0.13$, $P = 0.63$) nor root biomass ($\rho_{15} = -0.34$, $P = 0.2$) was significantly correlated with ^{15}N uptake in *D. macrolephara*. However, despite lower variation in ^{15}N acquisition in *T. triandra* (Table 1), both total biomass ($\rho_{15} = 0.60$, $P = 0.013$) and root biomass ($\rho_{15} = 0.62$, $P = 0.011$) were significantly correlated with ^{15}N uptake in this species.

Soil water content

Finally, soil volumetric water in laboratory pots displayed a significant species-by-clipping interaction ($F_{1,24} = 5.4$, $P = 0.03$). Pots containing clipped *T. triandra* had the highest volumetric water content, clipped *D. macrolephara* and unclipped *T. triandra* had intermediate and similar amounts of soil water, and unclipped *D. macrolephara* had the driest soil (Table 1). In addition, soil volumetric water content was a function of a soil texture-by-clipping interaction ($F_{1,24} = 4.2$, $P = 0.05$).

Table 2 Catena position (block), sand and clay content, soil gravimetric water content, grazing frequency, and above-ground (AG) biomass of grasses *D. macroblephara* and *T. triandra* at the study site in the mid-grass region of Serengeti National Park. Blocks were chosen to transect a soil catena gradient and biotic and abiotic factors that influence the abundance of dominant grasses

Block	Catena hillslope Position	Sand content (% sand) $n = 5$	Clay content (% clay) $n = 5$	March soil water (g H ₂ O 100 g ⁻¹) $n = 5$	May soil water (g H ₂ O 100 g ⁻¹) $n = 5$	Grazing frequency (% grazed) $n = 9$	<i>D. macroblephara</i> AG biomass (g m ⁻²) $n = 12$	<i>T. triandra</i> AG biomass (g m ⁻²) $n = 12$
1	Top	68.1 ± 2.8	13.7 ± 0.4	2.7 ± 0.1	8.1 ± 0.2	3.7 ± 0.1	41.4 ± 8.8	31.3 ± 4.4
2	Middle	52.9 ± 10.3	24.6 ± 1.7	5.3 ± 0.3	10.1 ± 4.0	17.3 ± 2.3	72.2 ± 7.2	23.4 ± 6.3
3	Bottom	44.5 ± 19.7	28.0 ± 9.8	6.7 ± 1.5	13.7 ± 12.3	24.1 ± 3.4	71.9 ± 9.0	27.2 ± 5.1
4	Middle	61.0 ± 51.1	25.0 ± 28.5	5.5 ± 2.2	5.6 ± 1.9	18.6 ± 1.8	41.9 ± 10.8	30.1 ± 6.5
5	Top	68.1 ± 9.6	15.2 ± 12.2	4.6 ± 0.1	5.4 ± 0.5	26.5 ± 2.3	101.3 ± 12.4	14.5 ± 4.0

Data are means (± SE) within blocks; n = sample size within blocks. See text for the methods used to determine sand content, gravimetric water content, grazing frequency and above-ground biomass; silt content (% silt) = 100 - (% sand + % clay).

Clipped plants grown in low sand had the highest volumetric water content (15.2 ± 2.2%), followed by clipped plants in high sand (9.6 ± 0.8%) and unclipped plants in low sand (8.6 ± 1.0%), which were not different by specific contrasts. Unclipped plants in high sand had the lowest soil moisture (6.7 ± 0.6%), except that they were not different from unclipped plants in low sand.

CATENA FIELD STUDY

Observational results

Long-term (> 40 years) average rainfall at the field site is 759 mm year⁻¹. Rainfall during the 3-month field experiment was typical for the mid-grass region of SNP, with a cumulative rainfall from 1 March to 1 June 2002 of 283.5 mm. There was a strong negative correlation between the above-ground biomass of *D. macroblephara* and *T. triandra* at five locations sampled across the catena gradient (Table 2; $\rho_4 = -0.95$, $P = 0.01$), although within-site variation in species biomass was considerable (Table 2). Variation in soil texture in the undisturbed grassland at the five blocks was consistent with theory describing soil catena gradients (Table 2): percentage sand was greatest at top hillslope positions and declined towards the bottom hillslope, while clay content was greatest in the bottom hillslope position and decreased towards top hillslopes. In both March and May, soil gravimetric water content was negatively related to soil percentage sand (March, $F_{1,23} = 76.6$, $r^2 = 0.77$, $P < 0.0001$; May, $F_{1,23} = 20.3$, $r^2 = 0.47$, $P = 0.0002$). Grazing frequency varied in a complex way across the catena gradient (Table 2); the highest and lowest grazing frequencies occurred at the top hillslope positions, while the bottom and middle hillslopes experienced high and intermediate grazing intensities, respectively. Across the catena, grazing frequency was the only variable associated with the above-ground biomass of the species, but in opposite directions. *D. macroblephara* abundance increased with grazing frequency (DM biomass = 2.1 × [%grazed] + 27.4, $F_{1,58} = 12.9$, $r^2 = 0.18$, $P = 0.0007$), while the relationship for *T. triandra* was negative, but relatively weak (TT biomass = -0.5 × [%grazed] + 34.6, $F_{1,58} = 2.81$, $r^2 = 0.05$, $P = 0.10$).

Growth and biomass

After 3 months, for both clipped and unclipped treatments, *D. macroblephara* final biomass (22.8 ± 1.2 g plant⁻¹) was greater than *T. triandra* (14.3 ± 1.3 g plant⁻¹, $F_{1,54} = 26.07$, $P = 0.007$); however, *T. triandra* final height (43.7 ± 2.1 cm) was greater than *D. macroblephara* (31.1 ± 2.1 cm, $F_{1,54} = 14.0$, $P = 0.02$). Soil texture had no effect on final biomass ($F_{2,54} = 2.73$, $P = 0.13$) or final height ($F_{2,54} = 0.58$, $P = 0.58$), and soil texture by species interactions were not significant for biomass ($F_{2,54} = 1.23$, $P = 0.34$) or final height ($F_{1,54} = 0.75$, $P = 0.5$). However, there was evidence that soil texture influenced biomass allocation differently in the two species, demonstrated by a marginally significant species-by-sand interaction on root biomass ($F_{1,54} = 2.56$, $P = 0.08$). Specific comparisons showed that root biomass was greater for *D. macroblephara* grown in the sandiest soils compared with those of the other treatments. As in the undisturbed soils, gravimetric water content decreased with sand content in the artificial subplots ($F_{1,43} = 118.1$, $r^2 = 0.73$, $P < 0.0001$).

¹⁵N uptake

The mean ¹⁵NO₃⁻ uptake in artificial plots for *D. macroblephara* was 445.2 ± 28.8 µg plant⁻¹ and 433.9 ± 29.3 µg plant⁻¹ for *T. triandra*, rates that were not significantly different ($F_{1,57} = 0.08$, $P = 0.78$). Simulated grazing increased ¹⁵NO₃⁻ uptake from 343.2 ± 28.8 µg plant⁻¹ in unclipped control plants to 535.9 ± 29.3 µg plant⁻¹ in clipped plants ($F_{1,57} = 22.0$, $P < 0.0001$). Clipping did not affect mean ¹⁵NO₃⁻ uptake of the two species differently ($F_{1,57} = 0.11$, $P = 0.74$). When unclipped plants were analysed with soil texture as a covariate, the slope of plant ¹⁵NO₃⁻ uptake versus percentage sand was not different from zero for *D. macroblephara* ($t_{14} = 0.97$, $P = 0.35$) or *T. triandra* ($t_{13} = 0.74$, $P = 0.47$). However, when clipped plants were analysed with sand as a covariate, the species had slopes that were significantly different from each other ($t_{14} = 3.98$, $P = 0.0013$). The regression of ¹⁵NO₃⁻ uptake versus percentage sand was positive for clipped *D. macroblephara* (Fig. 1A) and negative for clipped *T. triandra* (Fig. 1B). Rates of ¹⁵N uptake were not

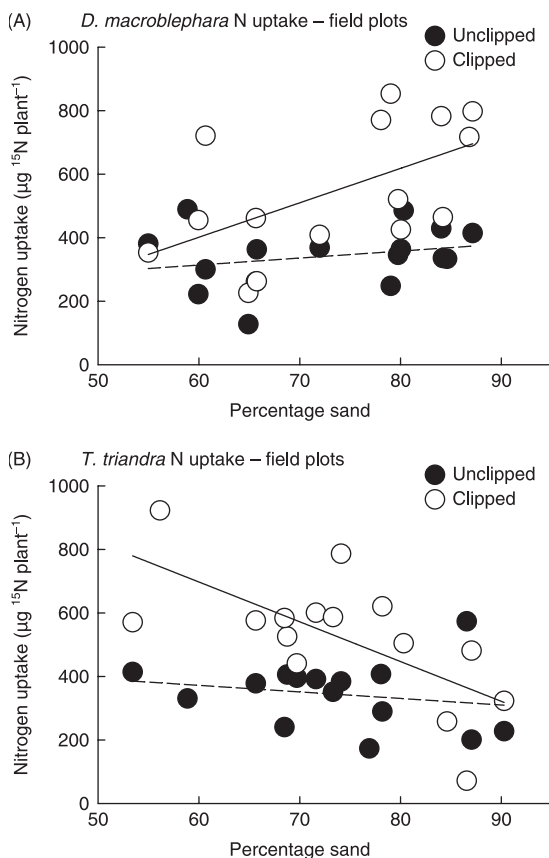


Fig. 1 Uptake of $K^{15}NO_3$ ($\mu g^{15}N\ plant^{-1}$) for individual plants of *Digitaria macroblephara* (A) and *Themeda triandra* (B) across soils of different texture (% sand) after 5 days (see text for methods). Slopes of clipped (unfilled circles) and unclipped (filled circles) plants were significantly different within both species and slopes of clipped plants differed significantly between species (see Results section for statistics). The regression equations were: *D. macroblephara* $^{15}NO_3$ uptake = $10.83 \times$ (percentage sand) $- 248.3$; $t_{13} = 2.45$, $r^2 = 0.32$, $P = 0.03$; and *T. triandra* $^{15}NO_3$ uptake = $-12.52 \times$ (percentage sand) $+ 1448.8$; $t_{13} = 3.25$, $r^2 = 0.45$, $P = 0.006$.

associated with variation in below-ground biomass of *D. macroblephara* ($\rho_{15} = 0.27$, $P = 0.14$) or *T. triandra* ($\rho_{15} = 0.08$, $P = 0.66$), or total biomass of *D. macroblephara* ($\rho_{15} = 0.04$, $P = 0.83$) or *T. triandra* ($\rho_{15} = -0.04$, $P = 0.83$).

Discussion

The results of our study suggest that dominant Serengeti grasses have distinct physiological responses to variation in soil texture and defoliation, and that the responses may be linked to their spatial distributions along soil catena gradients. *D. macroblephara* had positive physiological responses to defoliation and soils with more sand, including increased rates of photosynthesis, stimulation of key C_4 photosynthetic enzymes, and increased capacity for soil N assimilation. In contrast, when grown in soils with less sand, *T. triandra* had increased rates of photosynthesis, higher activities of at least photosynthetic and one N assimilating enzyme, and

greater final biomass. *T. triandra* also had increased N assimilation in the field when clipped, but only in soils with relatively lower sand content. While the species demonstrated notable differences in biomass allocation (i.e. root versus above-ground biomass) in response to clipping and soil texture, the total biomass response to soil texture was relatively weak (Table 1). A possible reason for this outcome may be that the experiments did not provide sufficient time for the physiological effects to translate into biomass differences. For example, the benefits of increased N assimilation may only fully manifest after the sustained dry season, when plants with greater nutrient reserves are more able to resist drought and thermal stress (Heckathorn *et al.* 1996) and can re-translocate N at the beginning of the wet season (Heckathorn & Delucia 1994), thus providing a physiological advantage over plants with lower N reserves. If the effects of soil texture and defoliation on N assimilation and physiological process compound over time as we expect, our results may provide a mechanistic link between soils, grazing herbivores, leaf-level physiological responses, and the observed distributions of dominant Serengeti grasses (Bell 1970, 1971; McNaughton 1983).

The removal of older, physiologically less active leaves and increased light interception to newly expanded leaves may explain increased activities of PPDK and PEPc in clipped *D. macroblephara* (Sheen 1999). Defoliation increases photosynthesis in many grasses (Detling *et al.* 1979; Caldwell *et al.* 1981; Painter & Detling 1981), including *T. triandra* (Wallace *et al.* 1984), and has been linked to the improved water status of Serengeti grasses (Toft *et al.* 1987), an effect consistent with the soil moisture results of this study. However, the stimulation of photosynthesis in clipped *T. triandra* and *D. macroblephara* appeared to have occurred for different reasons. Clipped *T. triandra* had greater stomatal conductance than unclipped plants, while the CO_2 concentration inside leaves was not different. Clipped *T. triandra* was not associated with greater enzyme activities of PPDK and PEPc, the enzymes responsible for carbon fixation in C_4 photosynthesis. In contrast, clipped *D. macroblephara* had greater stomatal conductance, higher leaf CO_2 concentrations, and was associated with higher activities of both PPDK and PEPc. Taken together, these results suggest that higher carbon assimilation in defoliated *T. triandra* resulted primarily from greater stomatal opening, while in *D. macroblephara* it was the result of greater stomatal opening coupled with higher biochemical activity of photosynthetic enzymes. This is consistent with previous studies of *T. triandra*, in which higher photosynthesis of clipped plants was attributed to higher stomatal conductance (Wallace *et al.* 1984).

Soil texture had opposite effects on the two species in several respects. In the field study, *D. macroblephara* invested more biomass in roots compared with *T. triandra* when grown in heavily textured soils. In the laboratory, sandy soils increased photosynthesis and PPDK activity in *D. macroblephara* but decreased photosynthesis and PPDK activity in *T. triandra*, which apparently resulted

in lower carboxylation efficiency and corresponding greater partial pressure of internal CO₂. Soil texture is an example of an 'indirect variable' (Ausin 1980; Ausin & Smith 1989) in that it influences plants by modifying other factors, the most obvious being alteration of water holding capacity (Jenny 1980; Fernandez-Illescas *et al.* 2001), a relationship that was observed in laboratory pots, artificial subplots and undisturbed catena soils. However, as we have no measure of soil water potential, we can only hypothesize that species were responding to differences in plant-available water. Estimates of soil matrix potentials across the catena hillslope, based on equations of Gupta & Larson (1979) using soil texture differences, provided general support for this idea. Average soil matrix potential of undisturbed soils at four of the five blocks were consistent with the results of soil water content: soils with the least volumetric water had the most negative matrix potentials while soils with more water had less negative soil matrix potentials (data not shown). This held at all hillslope positions except for block 1, where soil matrix potential was the least negative of the sites. However, as these are inexact estimates based on texture differences, future analyses will benefit from direct measures of plant-available water across Serengeti catenas.

One compelling aspect of this study was that after defoliation, species' ¹⁵N uptake rates in the field were opposite in response to soil texture. Grazing stimulates N uptake of other Serengeti plants (Ruess *et al.* 1983; McNaughton & Chapin 1985; Ruess 1988), but the dependence on soil texture is a novel finding. In a broad sense, this soil-grazer interaction is consistent with other mechanisms in grazing ecosystems that rely on plant-grazer feedbacks for maintenance of stable relationships (Augustine *et al.* 1998; Seabloom & Richards 2003). The results of the ¹⁵N uptake rates from the laboratory experiment confirmed that *D. macroblephara* grown in sandy soil had greater ¹⁵N assimilation when defoliated, but increased ¹⁵N uptake rates in low sand were not observed in *T. triandra*. The similarities and differences between the field and laboratory results are both revealing and ambiguous. For example, the results suggest that nutrient assimilation for *D. macroblephara* is not adversely affected by frequent defoliation and that grazer-induced N acquisition for *D. macroblephara* is consistent over a range of light conditions. In contrast, the discrepancy between the laboratory and field results for *T. triandra* may be due in part to low light conditions in the laboratory or a defoliation regime that was too frequent or intense. The second hypothesis is consistent with data that suggest *T. triandra* is intolerant to repeated defoliation (Hodgkinson *et al.* 1989; O'Connor 1994; Fynn & O'Connor 2000), but will require experimentation to resolve.

The differential uptake of ¹⁵N by plants grown in soils of different texture is most likely an example of an interaction between defoliation and water availability and the effect of water on soil N concentration. As an explanation of the different rates of N uptake by clipped *T. triandra* and *D. macroblephara* in a dry sandy vs.

moist clay soil, we propose the following mechanism, preceded by two important caveats. First, when equal amounts of aqueous inorganic N are added to soils, N in dry sandy soil is relatively concentrated and mobile compared with that in the moist, finer-textured soil. Secondly, *D. macroblephara* is more metabolically active than *T. triandra*, as shown by higher photosynthetic rates, enzyme activities and final biomass. We propose that the greater metabolic activity of *D. macroblephara* allows more rapid acquisition of the concentrated freely mobile N in the sandy soil before it is lost to leaching. Additionally, we propose that the concentrated N solution acts to increase the activity of N assimilation enzymes and thus biochemically increase N uptake in *D. macroblephara*. GS (Migge *et al.* 1996), NR (Campbell 1999) and PEPc (Pasqualini *et al.* 2001) are all regulated by the concentration of inorganic N. In contrast, the less metabolically active *T. triandra* can forage efficiently for relatively dilute N, but it does so slowly and in proportion to the quantity of foraging roots. In dry soils, *T. triandra* assimilates nutrients slowly, so lacks the physiological stimulation of enzyme activity induced by concentrated N. Thus, *T. triandra* benefits when N is retained and relatively immobile, as is the case in fine-textured soil.

There are three lines of support for this hypothesis. First, one study found that *T. triandra* photosynthesis was unaffected by different N concentrations (Wallace *et al.* 1985), suggesting that *T. triandra* may lack an up-regulation response altogether. Secondly, Ivans *et al.* (2003) found that the relatively fast growing tussock grass, *Agropyron desertorum*, begins N uptake immediately following ¹⁵N addition, while the slower growing native perennial shrub, *Artemisia tridentata*, did not respond to ¹⁵N until after 1 day, and did not reach similar uptake until after 2 days (Fig. 5 in Ivans *et al.* 2003). Thus, evidence exists that species differ in their timing of N uptake and less metabolically active species may respond more slowly than rapidly metabolizing species. The final line of evidence comes from the laboratory results. Activities of the four enzymes in *T. triandra* were significantly lower in high sand than in low sand treatments (Table 1). In opposition, activities of the four enzymes were higher in *D. macroblephara* grown in high sand (Table 1), which consistently had the lowest soil water. Finally, the relatively large increases in N assimilation for *D. macroblephara* could not be attributed to differences in total or root biomass, suggesting physiological regulation of N uptake. In contrast, the strong correlation between rates of ¹⁵N uptake and biomass for *T. triandra* in the laboratory, suggest that physiological changes in enzyme activity played little or no role in variation in N assimilation.

In the observational catena study, the trade-off in *T. triandra* and *D. macroblephara* biomass corroborates the finding that the species partition habitat along topographically variable soil gradients (Anderson & Talbot 1965; Bell 1970; Bell 1971). Across the catena, *D. macroblephara* was most abundant on a sandy hilltop

with the most negative soil water potential as estimated by texture properties (Gupta & Larson 1979) and the highest grazing frequency. *T. triandra* was most abundant at the site with the least negative soil water potential as estimated by soil texture (Gupta & Larson 1979) and its abundance across the catena was negatively related to grazing frequency. In fact, the abundances of the species along the catena gradient were correlated with grazing frequency in opposite directions, which matched their overall physiological response to grazing. In both the Masai-Mara and SNP, frequent and intense grazing increased *D. macroblephara* abundance and decreased *T. triandra* abundance (Heady 1966). In Omo National Park, Ethiopia, *D. macroblephara* increased in artificially defoliated plots but declined inside exclosures; apparently, *D. macroblephara* exploited canopy openings created by the loss of grazing intolerant species, while it was out-competed by grazing tolerant species when protected from grazing (Jacobs & Schloeder 2003). In South Africa, extensive demographic studies have demonstrated that rainfall has pronounced effects on the abundance of *T. triandra* (O'Connor 1993; O'Connor 1994; Fynn & O'Connor 2000). In periods of low rainfall, grazing exacerbates *T. triandra* mortality (O'Connor 1994), leading to its decline in the seed bank (O'Connor 1996). One study suggested that water availability had a stronger effect on *T. triandra* than grazing (Danckwerts & Stuart-Hill 1989), while other studies agree with the finding that defoliation has strong negative effects on *T. triandra* (Coughenour *et al.* 1985; Hodgkinson *et al.* 1989). O'Connor (1994) suggested that inconsistencies among studies of *T. triandra* may reflect an interaction between grazing and water availability, which is consistent with our hypothesis that soil texture effects are mediated through variation in soil moisture.

In regions of Serengeti where topographic variation and species co-occurrence is high, our data suggest that soil texture may play an important role in creating habitat heterogeneity by modifying water availability along catena gradients. When grazing is superimposed on the underlying template of soil heterogeneity that exists along catena gradients, our results suggest that defoliation and the interaction between water availability and defoliation strengthen opportunities for habitat partitioning and plant coexistence by promoting different physiological responses. Previous theories of plant coexistence in grazing ecosystems suggest that a trade-off between herbivore tolerance and grazing selectivity maintains coexistence in grasslands (Augustine & McNaughton 1998). Apparently, the shifting mosaic of grazing in the Serengeti contributes to species individualistic responses to the underlying patterns of spatial heterogeneity that are important for maintaining coexistence in species rich communities (Amarasekare 2003).

Acknowledgements

This study was possible because of help from E. Mayemba, Y. Byarugaba, C. Ruhanga, and M. Kibwe

in Serengeti. Thanks to M. Coughenour for use of a Land Rover, S. Heckathorn for laboratory equipment, R. B. McKane, J. Hamilton and M. Ritchie for discussions of the laboratory and field results, and to the Serengeti Ecological Monitoring Programme for providing rainfall data. Two anonymous reviewers improved the quality of the manuscript through conceptual and editorial contributions. This research was funded by an NSF doctoral fellowship to T.M.A. and NSF grant DEB-9903845 to S.J.M.

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Received 9 February 2006

revision accepted 24 April 2006

Handling Editor: Matthew Turnbull